OENOTHERA CYTOLOGY IN RELATION TO GENETICS[1]

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THE brilliant concurrence of cytology and experimental breeding in the solution of the fundamental problems of genetics has been one of the most striking characteristics of biological progress in the last quarter century. So mutually helpful have been these two directions of attack on genetical problems that we are ever more and more encouraged to carry over the discoveries in either of these fields as a basis for interpretation of observations and predictions of the results of further studies in the other field.

Of the two lines of work, cytological and genetical, the latter is far more delicate and effective, and we have a conception of the structure and behavior of chromosomes of great exactness and complexity as a result of genetical analysis, while the cytologist is as yet able to do little more than follow the movements of whole chromosomes as they spin out into delicate threads and pass into a state of invisibility, collect again into delicate filaments, contract into thicker threads usually appearing homogeneous in structure (though occasionally showing a split or double condition, and in some cases seemingly made up of bead-like masses of chromatin strung on a structural network), divide into chromosomes again, separate into equal halves to opposite poles of the mitotic figure,

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and go through the two highly characteristic meiotic divisions in gametogenesis.

Cytological research has demonstrated that eggs and sperms make approximately equal contributions to the chromatin content and chromosome number of the zygote, that the meiotic (maturation) divisions provide a mechanism which is adapted to produce exactly the observed normal Mendelian phenomena, that the two sexes of a dioecious organism usually possess visible chromosomal differences, that different species are frequently differentiated by different chromosome numbers, that special characteristics of the phenotype or exceptional behavior of the genotype in experimental breeding are, in a number of specific cases, associated with notable abnormalities in chromosome morphology or distribution, such as non-disjunction, fragmentation, cohesion, etc.   So decisive have been the results of such parallel studies on cytology and genetics, that it seems proper to seek a chromosomal explanation of all segregation phenomena (except such as can be referred to other discrete protoplasmic structures like the chloroplasts), and to find a genetical consequence of every observed cytological peculiarity which has to do with the number, arrangement or distribution of the chromosomes.

In Oenothera it is a particularly interesting fact that both the breeding results and cytological findings show noteworthy deviation from the conditions recognized as normal and characteristic in most other organisms.

The notable peculiarities of Oenothera breeding behavior consist in: (a) the true-breeding of demonstrably heterozygous parents; (b) the formation of "twin hybrids" in the F_1 of certain species crosses; (c) the production of relatively rare segregations in the F_2 and later generations from crosses; (d) the occurrence of unlike reciprocal hybrids; (e) the relative rarity of occurrence of inheritance ratios closely simulating the typical Mendelian ratios; (f) the occurrence of large degrees of pollen-, ovule- and seed-sterility, and (g) the ability of

practically every pair of species within the subgenus Onagra, however divergent in phenotypic characteristics, to cooperate in the production of fertile hybrids.

Besides these breeding phenomena in Oenothera which are unusual or absent in most other organisms, there may also occur the typical Mendelian and linkage phenomena which are nearly universal in the genetical behavior of other plants and animals.

The peculiarities of Oenothera cytology consist: (a) in the union of chromosomes end-to-end (telosynapsis) to form a so-called univalent spireme, which (b) subsequently breaks up into chromosomes, some or all of which may remain attached to each other, forming chains or circles, the number of chromosomes in each such chain or circle being usually characteristic for any particular species or mutant derivative, though several cases have been reported in which two or more different chromosomal arrangements have been found in forms exhibiting the same phenotype. (c) The chromosome chains or circles arrange themselves in the plane of the metaphase plate in the meiotic division, after which adjacent chromosomes are usually drawn to opposite poles of the spindle, giving the chain a zigzag appearance before the chromosomes become definitely separated from each other. (d) Rather frequent irregularities in the distribution of the chromosomes to the two poles have been found, chiefly of such a nature that two adjacent chromosomes go to the same pole instead of going to opposite poles.

In trying to correlate the peculiarities of Oenothera cytology with the peculiarities of Oenothera breeding behavior, Dr. Cleland, who has been largely responsible for the discoveries regarding the behavior of the chromosome circles during meiosis, has proposed an interpretation of the phenomena found at this phase, which has been accepted by a number of other workers who appear not to have made a thorough-going critique of the basic assumptions involved in Cleland's hypothesis. The essential feature of Cleland's interpretation is his assumption

that cohesion of the chromosomes of non-homologous
pairs into chains or circles, and the separation of alter-
nate chromosomes to opposite poles, constitute a mecha-
nism which will account for the extensive linkage of
genetic factors.   Factors which behave in a typical Men-
delian manner, only a few of which are yet known, are
supposed to be included in chromosomes which do not
remain attached to the chromosome chain or circle at
metaphase.

In identifying the chromosome circles as a mechanism
for the production of the linkages which constitute so
striking a characteristic of Oenothera genetics, Cleland
makes two assumptions which he ties together in such a
way as to make them seem almost like a single assump-
tion, namely, (a) "that the chromosomes are placed in
such a way in the circles that homologous ones are sepa-
rated to opposite poles" . . . and (b) " . . . the further
assumption that the chromosomes are not only arranged
into pairs but have definite positions within the pairs
also."   This second assumption means that chromosomes
of maternal and paternal origin alternate with each other
in the circle in a definitely fixed order, and that the draw-
ing of alternate chromosomes to opposite poles must,
therefore, carry all the maternal chromosomes of the
circle to one pole, and all the paternal chromosomes to
the other pole.   In this way would result the production
of only two kinds of eggs or two kinds of sperms with
respect to all the factors which happen to lie within the
chromosomes which enter into the chromosome-circle—
except, of course, when an irregularity of distribution
takes one or more paternal chromosomes to the otherwise
maternal pole and *vice versa*.   Such irregularities are
supposed to result in the phenomenon of crossing over.
Cleland argues at some length for the first of these two
assumptions and makes out a case so plausible that the
reader must keep a critical attitude if he is not to get the
impression that the strong case made out for the first
assumption carries with it the validity of the second as-
sumption also.

While Cleland has mostly avoided a specific statement that the convincing evidence which he adduces for the juxtaposition of homologous chromosomes in the circle supports in any way his second assumption that chromosomes of maternal and paternal origin alternate in regular order, he does state in one of his later papers (Cleland 1926a, p. 145) that "the constant presence of such configurations as these is as striking as it is unusual. It probably means that in the earlier stages in prophase the network has a definite structure, such that when it condenses to form the spiremes, the *individual*[2] chromosomes come out in the same way each time," and again (p. 149), "I have already emphasized the probability that the chromosomes are arranged within the circles in a definite order, so that homologues are as a rule separated to opposite poles. *This order probably applies not merely to the position of the pairs of homologues, but to that of individual chromosomes as well, so that each chromosome occupies a given position and does not ordinarily change position with any other chromosome, even with its homologue.*"[2]

In another paper Cleland (1926b, p. 65) makes the similar statement that "If it be true that each pair of homologous chromosomes is definitely placed within the circle, then it is most likely that *the same applies to each univalent chromosome also, and that each has its own place in relation to other chromosomes;*[2] and the circle which seems to be uniformly present throughout the species is uniformly constructed as well."

The acceptance of the same viewpoint by other students of Oenothera cytology may be indicated by two quotations from other recent papers: Thus, Håkansson says (1926, p. 258), "Vieles deutet darauf hin, dass bei Oenothera-Formen wo die Chromosomen während der Diakinese und nach der Auflösung der Kernmenbran in Form von Ketten oder Ringen zusammenhängen, *die Faktoren dazu tendieren gemeinsam vererbt zu werden, während freien Chromosomenpaare mit freier Mendel-*

[2] Italics are mine.

spaltung verknüpft sind.[3]   Der wahrscheinliche zytologische Mechanismus für diese Chromosomenkoppelung ist von Cleland entdeckt worden.'' Miss Sheffield (1927, p. 800) says similarly: ''It is reasonable to suppose that homologues are also adjacent when large rings persist into 'diakinesis'; *chromosomes of maternal and paternal origin would probably alternate. It seems to be more than likely that each chromosome has a definite position on the spireme, which it always assumes, the arrangement quite possibly persisting throughout the nuclei of the somatic tissues.*[3]

We need not allow the fact that Boedijn (1924), Emerson (1924) and Oehlkers (1926) have found somewhat divergent results, to discount the definite findings of Cleland, Håkansson and Sheffield, though it is important to remember, in each case of discrepancy of results, that the different workers have not investigated exactly the same material. To illustrate the importance of this I may cite the fact that Cleland has found in *Oe. franciscana sulfurea* from the cultures of Davis 12 chromosomes in a circle and one independent pair, while in *Oe. franciscana sulfurea* from my cultures he found ,no circle, but seven independent pairs.

Professor J. T. Illick, until recently of the University of Nanking, China, working in the biological laboratory of Princeton University in 1923–24, observed the phenomena of circle formation and distribution of alternate chromosomes to opposite poles, in full support of Cleland's discoveries, though in *Oe.* mut. *pervirens,* a form which has in most respects the same breeding behavior as *Oe. Lamarckiana,* he found two different configurations in different plants, sometimes a circle of twelve and an independent pair as in *Lamarckiana,* sometimes seven pairs as in *deserens.*[4]

Illick has been inclined, of course, to give his cytological observations genetical interpretations in accord with

[3] Italics are mine.

[4] Owing to the disturbances in China the completion of Illick's studies and publication of his paper have been delayed.

those of Cleland, but in a recent discussion with him of
the points which are presented in the present paper, he
has come to agree with me that there is a serious inade-
quacy in the evidence for Cleland's second hypothesis,
and that so far as the cytological evidence goes the proba-
bilities lie in a different direction from what has been
assumed by Cleland, Håkansson, Sheffield and others.

While giving full weight to the divergence of Oenothera
cytology from that of many other organisms, it does not
seem proper to go far beyond the actual evidence in order
to magnify the extent of this divergence.   In no organ-
ism, so far as I know, is there any evidence that maternal
and paternal chromosomes are oriented or distributed
in such manner as to indicate that their parental origin
has any influence whatever in determining their distribu-
tion or arrangement.   Cytological observations can give
little or no information on this point because there is
rarely, if ever, any way of identifying chromosomes of
maternal and of paternal origin except in hybridizations
between species so distinct from each other that the nor-
mal cooperation of the two sets of chromosomes in the
reproductive processes is fundamentally disturbed.   The
genetical evidence is decisive that in other organisms
paternal or maternal origin is a historical fact of no sig-
nificance whatever for the grouping of chromosomes in
the gametes, and this should make us very cautious about
the acceptance of such an assumption in regard to
Oenothera.   In fact, an assumption that there is an at-
traction between chromosomes of paternal origin for one
pole and of the maternal chromosomes for the opposite
pole (Cleland, 1926a, p. 150) deprives of all significance
the arrangement of chromosomes into circles as a deter-
mining feature in the assumed separation of chromo-
somes of paternal origin from those of maternal origin,
for the same attractive forces which are assumed to
orient the two unconnected circles of 6 and 8 chromo-
somes of *Oenothera biennis* would also serve to bring a
single disconnected pair into the metaphase plate with
the chromosome of paternal origin oriented toward the

putative paternal pole and the maternal chromosome toward the supposedly maternal pole.

In the absence of all evidence as to how the chromosomes arrange themselves following fertilization, may it not be reasonably supposed that homologous chromosomes become associated, but that the pairs of homologues thus formed may be oriented in any plane with reference to other pairs and that the connection between non-homologous chromosomes to form a continuous spireme occurs quite independently of the paternal or maternal source of the chromosomes? On this assumption the orientation of the chromosomes of each pair is a matter of chance, so that in following a series of spiremes from left to right any chromosome of maternal origin will lie to the left of its paternal homologue in 50 per cent. of the cases, on the average, and to the right in the remaining 50 per cent. Thus one spireme might have the arrangement A′ABB′CC′D′DEE′F′FG′G, another AA′BB′C′CD′ DEE′FF′G′G, and so on. If this should be the method of formation of the diploid spireme, the movement of successive chromosomes to opposite poles would result in the association of maternal and paternal chromosomes in the two daughter nuclei of the meiotic division, in a purely chance way, just as happens when all the chromosomes arrive at the metaphase plate of the meiotic spindle arranged in separate pairs of homologues.

Even if we assume with Cleland that the original arrangement in the spireme is a completely determinate one, AA′BB′CC′DD′EE′FF′GG′, it is doubtful whether the chromosomes would remain in the same order by the time they form the visible chromosome circles of the late meiotic prophase, for students of Oenothera cytology have assumed with plausibility that loops of spireme which are seen extending out from the synizetic knot at the time of ''second contraction'' and which usually show a break between chromosomes at their distal end, represent pairs of homologous chromosomes. Illick,[5] Cleland

[5] J. T. Illick, now of the Western Reserve Medical School, first discovered in the winter of 1923–24 the twisting and fusion of chromatin loops in such

and Håkansson have seen these loops twisted on themselves in such a manner as to suggest that opportunity is thus offered for an interchange of homologous chromosome parts which could result in the phenomenon of crossing over. As one examines the published figures of this phase of the meiotic process, it seems obvious that the loops show no tendency to arrange themselves in any single plane, but are oriented in an apparently random manner. If the loops can be twisted on themselves in such manner as to allow an exchange of parts, it should be even easier for the whole loop to turn through an angular distance of 180° or more, so that when the tangle again resolves itself into a simple circle or chain of telosynaptic chromosomes, the positions of the individual chromosomes making up the homologous pairs may conceivably be reversed with reference to each other as frequently as they would lie again in the same relative positions that they occupied previous to this second contraction stage.

While both the arrangement of the individual chromosomes in the original diploid spireme and the possible rearrangement at second contraction during meiosis seem to provide plausibly for independent arrangement of the homologous chromosomes within the single pairs, there is no reason to doubt the validity of the arguments of Cleland and others in support of the view that homologous chromosomes lie adjacent to each other in the "univalent" spireme, and that the zigzag arrangement of the chromosome circle at "diakinesis" represents the separation of homologous chromosomes to opposite poles.

manner as to lead to the conclusion that there is provided in the "second contraction" stage a mechanism which may produce crossing over between two loci in the same pair of homologous chromosomes. Soon thereafter Illick returned to his position in the University of Nanking, China, where he continued his Oenothera studies, but owing to the difficult political situation and an exciting but fortunately successful flight, in which he was separated for a long time from notes and MSS., the completion of his studies was considerably delayed and they have not yet been published, though now ready for publication. In the meantime Cleland (1926a, b) has confirmed the findings of Illick, and Håkansson (1926) has independently made the same discovery. See also p. 110.

We have thus a picture of the process of meiosis in Oenothera which gives practically the same results as in those organisms which show a parasynaptic arrangement of the chromosomes at meiotic metaphase.

One of the rather obvious difficulties for Cleland's hypothesis has been the observation of a considerably larger percentage of irregularities in the zigzag arrangement of the chromosome circles in early anaphase than can be correlated with observed mutational deviations in experimental breeding, particularly in such forms as *Oe. biennis* and *Oe. franciscana sulfurea* in which the mutation percentages are extremely low. He says (Cleland, 1926a, p. 156), "Approximately half of the sperm of normal chromosome number resulting from irregularities will have full complements of chromosomes and should prove functional. These will probably amount, therefore, to 3 or 4 per cent. of the total number of sperm, and consequently a number of plants in any given strain should show in some degree variations from the normal and uniform appearance." Cleland himself evidently senses this difficulty for he repeatedly urges that the number of observable mutations would be very low—limited presumably to cases of "maximum transference." He offers a sliding-scale method of production of mutations which does not harmonize well with the experiences of Oenothera breeders when he says: "The individuals resulting from the union of the modified complexes with normal gametes will vary from the normal in a degree depending upon the number of chromosomes which have crossed from one complex to the other. *In the case of maximum transference the resulting individual may perhaps be so different that it will be regarded as a mutant.*"[6] The same untenable idea is expressed by Miss Sheffield (1927, p. 803), who says, "Such irregularity would result in aberrations or, *if sufficient characters are involved,*[6] in mutations."

Miss Sheffield (1927, p. 801) points out that some of the observed irregularity in the zigzag arrangement of the

[6] Italics are mine.

chromosomes may not result in a change of constitution
of the putative paternal or maternal complexes, but may,
indeed, *serve to correct a previously acquired irregu-
larity*[6] in the otherwise definite alternation of paternal
and maternal chromosomes in the original diploid
spireme.   She says, "It is just possible that an abnor-
mality occurred in the order of the chromosomes on the
spireme and that this irregularity is being rectified, so
giving the erroneous idea that an abnormality is occur-
ring at anaphase."   In this assumption that an irregu-
larity in arrangement of chromosomes in the spireme
might have occurred, Sheffield comes very near antici-
pating the viewpoint presented in this paper.   From the
standpoint of my conception of the composition of the
spireme, a deviation in the orderly zigzag arrangement
such that two adjacent chromosomes are drawn to the
same pole would probably have no genetical significance
whatever when it is balanced by two other adjacent
chromosomes going to the opposite pole, since the two
adjacent chromosomes involved in each such irregularity
would be most likely non-homologous chromosomes, and
the normal separation of homologous pairs would not be
in any way disturbed by such irregularity nor would the
normal chance association of chromosomes of maternal
and paternal origin in the daughter nuclei be in any way
affected by it.

From the point of view here set forth, that a univalent
spireme of the Oenothera type is made up of a succession
of pairs of homologous chromosomes in which the order
of the individual chromosomes with respect to their
maternal or paternal origin is indeterminate, the occur-
rence of chromosome circles in the meiotic prophase and
metaphase must be looked upon as having no very funda-
mental genetical significance.   There are certain other
facts which seem to be in accord with this conclusion.
The cytological investigations have discovered in the dif-
ferent species and mutants of Oenothera practically
every possible association of circles and free pairs, except
that no circle of 10 associated with 2 free pairs has yet

been reported. (This arrangement also will surely be found sooner or later.) If we arrange the forms in order of the number of chromosomes included in the circles we have the following series:

> Circle of 14, *Oe. muricata* (Cleland, 1925); *nova-scotiae, eriensis* (Sheffield, 1927)
>
> Two circles of 6 and 8, *Oe. biennis* (Cleland, 1923, 1926; Emerson, 1924)
>
> Circle of 12 or 14, *Oe. suaveolens, Cockerelli, strigosa* (Oehlkers, 1926)
>
> Circle of 2, *Oe. Lamarckiana* (Cleland, 1925; Håkansson, 1926), *franciscana sulfurea* (Cleland, 1924), *planifolia, flavescens, gigantea* (diploid) (Håkansson, 1926), *ammophila* (Sheffield, 1927)
>
> Circles variable, *Oe. Agari* (Sheffield, 1927)
>
> Circle of 8, *Oe. rubricalyx* ("Afterglow") (Cleland, 1925)
>
> Circle of 6, *Oe. rubrinervis* (Cleland, 1925), *rubrisepala* (Håkansson, 1926), *rubricalyx* (Sheffield, 1927)
>
> Circle of 4, *Oe. franciscana* (Cleland, 1922)
>
> No circle, *Oe. grandiflora* (Davis, 1909), *Hookeri* (Schwemmle, 1924), *Lamarckiana* (Boedijn, 1924), *blandina, deserens* (Cleland, 1925) *latifrons*,[7] *franciscana sulfurea*.[7]

There is no suggestion of a graded series of differences in the genetical behavior of the foregoing species except in *Oe. Lamarckiana* and its derivatives. Each species seems to have its own special breeding behavior, dependent upon its particular genotypic constitution with reference to the presence or absence of lethal factors, and whether these are gamete lethals or zygote lethals. Admittedly, knowledge of the breeding behavior of most of these species is still too meager, and it may be that when all of them have been studied as long and intensively as *Oe. Lamarckiana* and its mutant derivatives have been studied, correlations between circle formation and genetical behavior will be seen that are not now apparent. As matters stand at present, we see *Oe. Lamarckiana* with its relatively high mutability and a breeding behavior seemingly dominated by the presence of two balanced zygote lethals which occur in no other known species, having the same arrangement of chromosomes as *Oe. franciscana sulfurea* and *Oe. ammophila* in which the breeding results show no semblance with those of *Lamarckiana;* and *Oe. franciscana* with a circle of four,

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[7] Determined by Cleland but not yet published, so far as I know.

one of the least variable species known, may be compared
with *rubrinervis,* which is most nearly like it in chromo-
some configuration, having a circle of six, but which splits
in every generation in approximately a 2:1 ratio of
rubrinervis and *deserens* besides producing occasional
mutations.   The assumption that the chromosomes are
being held together in an end-to-end arrangement because
of their own heterozygosity seems to be of particularly
doubtful validity in the case of *Oe. franciscana* in which
the circle is later resolved into two pairs, but in which
no hereditary variability has ever been reported.

According to Cleland's conception of the constitution
of the spireme, the zigzag arrangement of the chromo-
some circles in early anaphase makes the entire circle
the physical basis of a single linkage group; if the view
of the constitution of the spireme set forth in this paper
is correct, the zigzag arrangement is still of equal impor-
tance because of the assurance it gives that the chromo-
somes of each homologous pair are being separated in a
definite and orderly way at meiosis, thus making each
pair just as effective a basis for a linkage group as if the
homologous chromosomes were assembled parasynapti-
cally instead of telosynaptically.   The discovery of a
third linkage group, with three linked factors now known
(Shull, 1925, 1926, 1927, 1928) in a species which has only
one free pair of chromosomes besides the circle of 12,
appears to me to be strong evidence in favor of the view
that each pair of chromosomes may represent a separate
linkage group, whether it be included in the circle or
separated from it.

The fact that most of the known genetic factors in
Oenothera are included in linkage group I (Shull, 1921,
1923a, b) has seemed to some a strong argument in favor
of the view that the chromosome circle, instead of a single
pair of chromosomes, constitutes the basis of this linkage
group, but this argument should not be too heavily
weighted, since the number of investigated factors is
actually not large (compared, for instance, with 150 or
more known factors in a single pair of chromosomes in

Drosophila), and since it is admitted (Shull, 1923, Cleland, 1926a, p. 148) that the presence of lethal factors in a given linkage group has favored the accumulation of mutant factors in the same linkage group, which might otherwise have been eliminated.

A second argument against the acceptance of the single chromosome pair as the basis of a linkage group in Oenothera was offered by Gates (1922), who insisted that no mechanism exists for the production of the observed crossing over. The discovery of Illick, Cleland and Håkansson, that opportunity is presented at the time of second contraction for the interchange of parts of homologous chromosomes, clearly cancels the validity of Gates's argument.

As indicated in the footnote on page 104, the condition at "second contraction" which accounts reasonably for crossing over between the loci of a single chromosome pair has been known at Princeton since the winter of 1923–24, owing to the cytological studies of Illick. That there can be no question as to the validity of Illick's observations and conclusions is indicated from the following quotations from papers of Cleland and Håkansson: Cleland (1926a, p. 123) says, "At or near the time of maximum contraction, one sometimes notes that the sides of the radiating loops appear to be twisted around each other (Fig. 5). This approach to a strepsinema condition might result occasionally in the kind of chromatin exchange known as crossing over"; and again (p. 157), "In some cells in late second contraction I have noticed that the appressed sides in the case of one or more of the peripheral loops have apparently become twisted about each other in a way that might conceivably result in the exchange of chromatin particles." Håkansson (1926), working quite independently of Cleland, has simultaneously announced similar observations and conclusions. He says (p. 262):

. . . in anderen Fällen werden Chromosomen an den beiden Schenkeln der Schleife gebildet, und wenn diese einander berühren (Fig. 1), sieht es aus, als ob ein Chromosomenpaar durch Spaltung des Fadens entstanden sei, was

auf Parasyndese hindeuten würde. . . . Es sei erwähnt, dass wenn die beiden Schenkel der Schleife so nahe neben einander liegen, und sie diesfalls homologe Chromosomen representieren, eine Möglichkeit für Crossing over zwischen ihnen vorhanden sein kann, trotzdem dass eine metasyndetische Chromosomenpaarung stattgefunden hat. . . . Auch ist die Möglichkeit vorhanden, dass beim Einschnüren des Pachynemafadens ein Chromosomenstück von einem Chromosom zu einem anderen gelangt. [And again (p. 287):] Ungeachtet der metasyndetischen Chromosomenpaarung gibt es jedoch während dem ''Second Contraction,'' bei Formen wo diese stark ist, Möglichkeit zum Substanzaustausch.

In his English summary Håkansson says (p. 300): "The chromosomes synapse metasyndetically, constrictions appearing in the thick pachynema thread which is thus transformed into a ring of chromosomes attached end-to-end. There is, however, a possibility that crossing-over may occur in the second contraction."

Admittedly, the conception which is presented in this paper, namely, that the spireme in Oenothera is composed of chromosomes arranged in homologous pairs, with the order of arrangement of the maternal and paternal member of each pair indeterminate, is just as hypothetical as Cleland's conception, which assumes that maternal and paternal chromosomes alternate with each other in a definitely fixed order. Opinions may well differ at present as to the relative probabilities in favor of the one or the other of these two hypotheses. Fortunately a decision between them seems capable of being attained by experimental methods, and should be most quickly achieved through a close cooperation between the experimental breeder and the cytologist.

Summary

1. In attempting to relate the chromosome circles which are observed in many species of Oenothera, to the extensive linkages which characterize this group of plants, Cleland makes two assumptions: (a) that homologous chromosomes are adjacent to each other in the circles, and (b) that chromosomes of maternal and paternal origin alternate with each other in a definitely fixed order.

2. The evidence cited by Cleland and others for the first of these theses is convincing, but not for the second.

3. In the absence of evidence regarding the arrangement of the chromosomes in the original diploid spireme following fertilization, it seems reasonable to assume that homologous chromosomes become associated in pairs which may lie in any plane with reference to adjacent pairs, and that when the spireme is formed, the maternal or paternal origin of the chromosomes has no influence in determining the order of arrangement of the members of each pair of homologues.

4. Even if it be assumed that the order of succession of maternal and paternal chromosomes in the original spireme is definite, they need not remain in the same order until the chromosome circles emerge in late meiotic prophase, since the looping of the chromosomes during ''second contraction'' provides for possible reversals in the positions of the homologous chromosomes.

5. A spireme in which each pair of homologues remains associated, but in which the order of maternal and paternal chromosomes is indeterminate, provides for the normal separation of homologues when adjacent chromosomes move to opposite poles of the meiotic spindle, and such separation is probably achieved even when adjacent chromosomes move to the same pole, if two other adjacent chromosomes in some other part of the circle move together to the other pole.

6. It also provides for a chance assortment at each pole, of chromosomes of maternal and paternal origin, just as is known to take place when the arrangement of the chromosomes is parasynaptic.

7. This being the case, there may be as many independent linkage groups as there are haploid chromosomes regardless of the grouping of the chromosomes into circles.   The discovery of three linkage groups in *Oe. Lamarckiana*, in which the chromosomes are associated in a circle of twelve and one pair, is favorable to this view.

8. The telosynaptic arrangement of the chromosomes and their persistent cohesion into chains or circles, therefore, probably have no very fundamental genetical significance.

9. Irregularities in distribution of the chromosomes of the circles at anaphase are more frequent in such stable species as *Oe. biennis, Oe. muricata* and *Oe. franciscana sulfurea* than the number of visible genetical variations would lead one to expect if each such irregularity resulted in the exchange of genetically dissimilar chromosomes from one parental "complex" to the other.

10. Different species having like arrangement of the chromosome circles do not agree with one another in their genetical behavior, but seem to differ from each other rather on the basis of the number and nature of the lethal factors they possess.

11. Studies of Illick, Cleland and Håkansson have shown that at the time of second contraction in meiotic prophase, chromatin loops which probably represent pairs of homologous chromosomes show twisting and fusion which provide the possibility of transfer of chromatin particles or of whole sections of the chromosome from one homologue to the other, thus providing for crossing over within the linkage groups, notwithstanding the fact that the chromosomes are arranged telosynaptically. This sets aside Gates's (1922) contention that no mechanism exists for the production of crossing over in Oenothera.

12. The conception of the spireme here presented is just as hypothetical as is Cleland's view. Fortunately a decision between the two views seems capable of experimental attainment, especially through close cooperation between the experimental breeder and the cytologist.

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THE POSSIBLE MODIFICATION OF THE RESPONSE OF THE WILD TYPE TO RECURRENT MUTATIONS

DR. R. A. FISHER

ROTHAMSTED EXPERIMENTAL STATION

1. *Gene Mutations*

THE very extensive series of gene mutations observed in *Drosophila melanogaster* and allied species is at present by far the most ample basis available for the study of these changes; nevertheless the nature of the mutant genes found in other groups may be cited as indicating that the outstanding characteristics of the *Drosophila* mutations may be characteristic of gene mutations in general. By far the most numerous classes are either (i) viable when homozygous, and completely or almost completely recessive, or (ii) lethal when homozygous. Of the latter class many are apparently completely recessive in their action, while in others the heterozygote is distinguishable from the wild type, showing so-called dominant characters. A third and much less numerous class are non-lethal dominants, in which, however, the dominance is usually incomplete.

I have listed, with some uncertainty, the non-lethal mutations of *Drosophila melanogaster* having visible effect, from the article of Morgan, Bridges and Sturtevant in *Bibliographica Genetica* (1), with the result shown in the following table:

	Recessive	Incomplete dominant	Complete dominant	Total
Autosomal	130	9	0	139
Sex-linked	78	4	0	82

The two alternative statements, that the mutant type is generally recessive, or that the wild type is generally dominant, are formally equivalent; nevertheless, the latter statement is to be preferred, in view of the behavior

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of series of multiple allelomorphs, of which *Drosophila* furnishes several examples, which have been admirably paralleled in the albino series in rodents. In these cases it is found that while the wild type is clearly dominant to the mutant allelomorphs, yet the heterozygote of two mutant allelomorphs is intermediate between them (1. p. 34). The mutant allelomorphs show little or no dominance *inter se,* although it has been demonstrated that one can arise as a mutation from another. This group of observations suggests, therefore, that it is rather a peculiarity of the wild type to be generally dominant than a peculiarity of the mutant to be recessive to the type from which it arose.

This feature in the behavior of multiple allelomorphs appears to offer a serious difficulty to the theory that the evolutionary adaptation of specific forms has taken place by the occasional and gradual replacement by mutant genes of the allelomorphic wild type genes from which they arose. For, in their dominance, the wild type genes appear to be clearly of a different nature from the mutant genes which arise from them. This difficulty will lose its force if it appears that there is a tendency always at work in nature which modifies the response of the organism to each mutant gene in such a way that the wild type tends to *become* dominant. It is the purpose of the present paper to examine this possibility.

2. *The Mutations Available for Study Are Probably Recurrent*

Enormous as is the number of experimental flies among which the *Drosophila* mutants have occurred, this number must fall short a thousandfold of the number produced in the wild state even in a single year. Unless great differences in the mutation rates are postulated, ascribable to the artificial environment, it follows that, when any mutation has occurred repeatedly under observation, we may be confident that this mutation occurs in nature with a frequency, at a moderate estimate, of some-

thing like a thousand times annually. As to the period during which this process has continued, we may best judge by the fact that homologous mutations have been proved to occur in different species. This fact gives us only a lower limit, since more distantly related flies have not yet been sufficiently examined, and, where crossing can not be practiced homology can not absolutely be proved. The occurrence of albino mutants in very different mammals suggests at least that this mutation has been occurring since an early stage in the differentiation of the group.

With respect to the majority of the *Drosophila* mutations which have occurred only once under observation, it is also probable that each has occurred with enormous total frequency during the history of the species, for this would be the case even if the frequency of mutation was only one in 10^{12}, while a mutation occurring so rarely as this would have a very small chance of being detected in an experimental sample, which does not much exceed twenty-five millions; and we should expect to have detected few or no such mutants unless they were an enormously numerous class widely sundered in mutation frequency from the smaller group which are known to be recurrent. Such a view seems extremely improbable in contrast with the view that the majority of the mutations which we can hope to observe in experimental cultures, unless they can be ascribed to cultural conditions, are mutations of which the species has at least very extensive previous experience.

3. *Modification of the Reaction in Experimental Cultures*

That such experience is capable to leading to a modification of the reaction of the species to the mutant gene is shown by an important fact which has now been repeatedly verified. Mutant types which have been kept as stock for several generations have been observed to show their mutant peculiarities in a materially lower degree than at their first appearance. This remarkable phe-

nomenon has been ascribed to the differential survival of
the flies in the stock bottles, those having a hereditary
composition which gives a mild reaction to the mutant
gene having been more viable than those with a different
hereditary equipment which reacted more strongly.
That this view is correct is shown by the crucial experi-
ment of mating these modified mutants to unrelated non-
mutant stock, and extracting mutant flies from the off-
spring by inbreeding. It is found that this procedure
restores to the mutant type much or all of its former pro-
nounced character. The experiments demonstrate two
points on which we shall rely: (i) that it is not the mutant
gene which is modified but other hereditary factors which
influence the reaction of the organism to the mutant gene;
(ii) that those modifying factors which influence the via-
bility of the mutant type do so in such a way as to influ-
ence *pari passu* its external characteristics.

The first of these points is essential to any theory of
the modification of the response of the organism to a par-
ticular mutation, for the previous mutant genes must
sooner or later all become extinct, and any mutant gene
observed to occur in culture is in any case unmodified;
whereas, since mutant individuals, heterozygous or
homozygous, will leave a certain proportion of non-
mutant descendants, the selection of modifications in the
mutant types will affect the frequency of the correspond-
ing modifiers in the population at large. The second
point is of importance in showing that natural selection
in such types, although based solely upon survival in
competitive conditions, may be expected to modify those
external characters, by the appearance of which, in the
heterozygote, we judge which of the homozygous forms
is to be called dominant.

4. *Selection of Modifiers in Natural Conditions*

In natural conditions we may assume that any type
materially different from the wild form, such as the
homozygote mutant, or the heterozygote if the mutation

is not almost completely recessive, will be at a disadvantage. For purposes of calculation we may express the whole of this disadvantage as though it depended merely on viability, that is, on the frequency with which the fertilized ovum survives to reproduce, any disadvantage in such characters as mating instincts, fertility, or instincts governing oviposition being expressed as an equivalent failure of viability. For simplicity we may also assume that mating is at random, at least in the sense that the choice of a mate is not affected by the gene in question. In these circumstances it is obvious that if the mutant gene is kept rare by counter-selection, the homozygous mutant type will always be very much rarer than the heterozygote. The actual frequency of each type will depend on the intensity of the counter-selection, the frequency of mutation being regarded as constant. If the heterozygote is in any appreciable degree less viable than the wild form, any counter-selection, exerted against the homozygote mutant, even complete lethality, will be of no importance in determining the frequency ratio of the genes at which the opposing influences of mutation and selection come to an equilibrium. Thus if one wild type gene in a million mutates in each generation, and v stand for the viability of the heterozygote as compared to the wild type, the relative frequencies of the three possible types will be, homozygote mutant, p^2, heterozygote, $2p$, wild type, 1; where p is given approximately by the equation

$$p = \frac{10^{-6}}{1 - v}$$

or, in general, if k stand for the mutation rate, supposed small, by

$$p\,(1 - v) = k$$

At one extreme, if v is a small fraction, p is nearly equal to k, and the heterozygotes will occur two million times as frequently as the mutant homozygotes; at the other extreme if $v = 0.99$, so that the heterozygote is only at a one per cent. disadvantage, the heterozygotes will still be twenty thousand times the more frequent.

These very high ratios justify the conclusion that if the *heterozygote is at any appreciable disadvantage compared to the wild type, it will be so enormously more frequent than the homozygote that any selection of modifiers which is in progress will be determined by the reaction of the heterozygote.*

Two other circumstances serve to increase the disproportion of the selective effects. In the first place, the efficacy of the selection in modifying the characteristics of the species depends not only upon the frequency of the individuals selected, but upon their chance of leaving a remote posterity. In fact we need to evaluate not the relative numbers of the two types in any one generation but the proportions they represent of the total ancestry of a distant subsequent generation. Evidently, if, as is to be anticipated, the viability of the homozygous mutant is lower than that of the heterozygote, the latter will count for more in future generations, and even if the two types had equal viability, the heterozygote is still at an advantage, for mated to a wild type only half his offspring will be heterozygous, while in similar case all the offspring of the heterozygote will be equally handicapped.

This point becomes of importance with sex-linked factors when the mutant type males and the heterozygous females do not differ greatly in frequency, but may differ greatly in viability, with the result that the latter may occur much more frequently in the ancestry of the existing wild population.

In the second place there is some evidence that the heterozygote is naturally more modifiable than are the homozygous types. How much weight should be given to this consideration, it is not easy to say.

The fraction of the ancestry of future generations ascribable to heterozygotes, though greatly exceeding that due to mutant homozygotes, is still exceedingly small, it may be expressed approximately as

$$\frac{2p(p-k)}{p+k} = \frac{2kv}{(1-v)(2-v)}$$

and this represents the ratio of the rate of change to be expected in nature to the rate of change which could be effected by exposing a population composed entirely of heterozygotes to selection of the same intensity. This fraction is, of course, extremely small; if k is 10^{-6}, its value is only a trifle greater if $v = \frac{1}{2}$, rising to about one five thousandth if v is as high as 0.99. Nevertheless, considering the ratio of the periods of time available it seems not impossible, but rather probable, that the reaction of the wild type to the heterozygous phase of a recurrent mutation has in some cases at least been modified to an appreciable extent.

The efficacy of selection in modifying the heterozygous type of an imperfectly dominant mutation is open to experimental verification, and some idea could thereby be obtained as to the possible speed of the process. There is one respect, however, in which it will certainly be difficult to allow the experimental conditions to do full justice to the natural conditions which they simulate. The intensity of selection acting on any particular modifying factor would have to be perhaps 100,000 times greater than under nature. It would be difficult to prevent selections of this intensity from depleting the supply of modificatory variance available in any small experimental group of flies, and in consequence the efficacy of selection, if the experiment were prolonged beyond a few generations would fall off from a cause which would have no parallel in the natural evolution of the species. The maximal rate of progress observed might be expected to supply a better estimate than the average over a considerable period; though the latter doubtless would be easier to observe with precision.

It will be noticed that the fraction calculated above decreases very rapidly as $v \rightarrow 0$, and also increases very rapidly as $v \rightarrow 1$. The early stages in the improvement of the heterozygote will necessarily be retarded very much more severely than the later stages, which may be expected to be passed through quite rapidly in compari-

son.   Heterozygotes, the viability of which is much impaired will thus have made no appreciable progress by the time that others less heavily handicapped at the start will have become completely normal, in viability at least, and presumably also in appearance.   Whatever were the relative frequencies of dominants, recessive and intermediates of various grades among mutations at their first occurrence, we should expect, if the above selective process has had time to produce great effects, to find that the greater number of recurrent mutations had become completely recessive.   And this is what appears to be the case, in *Drosophila* and, with some exceptions, in other organisms generally.   A certain number of cases should, however, be found, if the survey is carried sufficiently far, in which the reaction of the organism to the mutant gene is nearly in its original condition, either because the mutation is really of comparatively recent origin, or because it affects the chances of life so seriously, in the wild environment, that no appreciable progress has been made in its modification.

5. *Modification of the homozygote*

Until the heterozygote has attained practically normal viability, there will be no appreciable tendency for the reaction of the organism to the homozygous mutant to be modified, except in so far as some such an effect has already been produced by the selective modification of the heterozygote.   The last stages in the approach to normal viability will be carried out comparatively rapidly, for the intensity of selection itself varies inversely to $1 - v$; during these final stages p increases very rapidly so that the numbers exposed to selective action not only of the heterozygote, but of the mutant homozygote will be very greatly increased.

If a stage is reached at which the heterozygote survives absolutely as well as the wild type, the frequency of the mutant gene will begin to depend upon the viability of the homozygote; if w stand for the viability of this geno-

type, the condition of equilibrium corresponding to equation I is

$$p^2(1-w) = k, \hspace{4cm} \text{II}$$

while the fraction of the ancestry of a remote posterity which in any one generation are homozygotes will be

$$\frac{kw}{1-w}$$

If w is small it is seen that the rate of progress at this stage is the same as that of a heterozygote of the same viability; so long at least as this progress is so slow that the rate of increase in the number of mutant genes is small compared to the rate of mutation. It is only necessary to note that this process can not commence until the viability of the heterozygote is, within a very minute fraction, equal to that of the wild type; and that if w is not small we are approaching a condition in which the mutation rate is not balanced by counterselection, and in which other considerations, such as reverse mutations, cease to be negligible.

It is clear, however, that a persistent mutation in which even the homozygote has not too bad a chance of survival, the homozygote may follow in the footsteps of the heterozygote, and become indistinguishable from the wild form. Such mutations might in fact leave no trace for genetic research to reveal. The sex chromosome provides the best opportunities for such a complete obliteration.

6. *The pristine character of the reaction of an organism to a gene mutation*

We should, on the view developed above, expect to find the appearance of the heterozygote of two alternative genes, in its original condition, only in somewhat exceptional cases.

(i) The occurrence of a mutation which is really comparatively new in the history of the species;
(ii) Mutations in which even the heterozygote has had from the first an exceedingly small chance of survival;

(iii) Different mutations of the same gene, which can be brought together experimentally, but which can seldom have come together in nature;

(iv) Specific modifiers producing no external effect save in the presence of mutant genes in other loci.

It can scarcely be hoped that more than a very small proportion of mutations occurring in experimental cultures can belong to the first class. Clearly, Muller's success, in multiplying the mutation frequency by means of X-rays, gives the best hope in the direction of securing mutants which have seldom occurred before. If, on the other hand, the adaptive modification of domestic varieties of animals and plants has taken place by the accumulation of favorable mutations, a certain number of comparatively new factors should be found to distinguish highly selected varieties from more primitive varieties of the same species; though even here it is not impossible that use may often have been made of old mutations which had repeatedly occurred and been rejected as unfavorable in the wild condition.

It seems possible to regard the dominant mutations in *Drosophila* as, for the most part, cases in which the chances of life of the heterozygote in the wild state are so low that no appreciable modification has taken place. If this is the case it may be significant that the dominance is usually, if not always, conspicuously incomplete, the homozygote being either lethal in its action, or exhibiting a more pronounced effect than the heterozygote.

The heterozygotes of allelomorphic mutants show a strong tendency to lack dominance altogether. Wright's quantitative study (2) of the albino series in guinea-pigs shows this effect very clearly. Morgan, Bridges and Sturtevant (1) give the rule as general for *Drosophila melanogaster,* though possibly there are exceptions. These certainly exist, though they are rare, in other organisms; thus the Himalayan allelomorph of the albino series in rabbits is dominant to the complete albino. Instances are rather numerous in the literature of one

allelomorph being spoken of as dominant to a second, when in reality the heterozygote is clearly intermediate; this loose use of the term makes it difficult to summarize the evidence of the several multiple allelomorph series found in mammals. Such evidence is, however, in any case on a different footing from that drawn from the *Drosophila* mutations, for some of the genes involved in domesticated varieties have been exposed in large numbers to the selection of fanciers, and in some cases it even appears that the heterozygote has been particularly selected by the fancier to conform to his ideal.

In opposition to the view suggested by the above classes of facts that complete dominance is not characteristic of the reaction of the wild type to a mutation at its first appearance, the specific modifiers seem often to show complete dominance. Thus of 6 modifiers of eosin eye color which have no visible effect on the wild fly, Bridges (3) finds only one with intermediate heterozygote, the remaining five being ordinary recessives. Since it is not easy to see how dominance in such cases could be brought about by selection in stocks in which the eosin eye only occurred as a rare mutant, such cases as these seem to indicate that for the mutant condition to be completely recessive to the wild type is a frequent occurrence from the start, though possibly less frequent than the proportion of recessive mutants observed in *Drosophila* would suggest. A comprehensive summary of the behavior of specific modifiers from this point of view would be of very great interest.

Finally, the possibility discussed above that the dominance of the wild type is in some cases a product of evolutionary change in the manner outlined throws no light upon such a case as is presented in poultry, where a majority of the genes which distinguish domesticated races from the wild type appear to be non-lethal dominants.

7. *Summary*

The consideration that the wild population is immensely larger than the numbers bred experimentally

even in *Drosophila,* and that the duration of the species is still more disproportionate to the experimental periods, leads to the conclusion that the majority of the mutations which we can hope to observe in cultures must have occurred previously with very great total frequency in the wild species.

The reaction of the wild type to mutations is known in many cases to be capable of a somewhat rapid modification in experimental conditions, by the selection through differential viability of factors capable of modifying this response.

It may be calculated that with mutation rates of the order of one in a million the corresponding selection in the state of nature, though extremely slow, can not safely be neglected in the case of the heterozygotes.

The observed behavior of multiple allelomorphs largely supports, though that of specific modifiers seems to oppose, the view that complete dominance generally may be regarded as a product of such selective modification.

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TAXI WINGS, A NEW USEFUL III CHROMOSOME MUTANT IN DROSOPHILA MELANOGASTER

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A LARGE number of new mutant characters in *Drosophila melanogaster* are annually being discovered and studied due to the universal use of this fly in biological laboratories and consequently the great number of individuals subjected to scrutiny. Among the numerous new mutants there appear a few possessing characteristics which make them useful in further genetic studies. Taxi is a recessive character of this type. It is easily and readily recognized, is fully fertile and almost as hardy as the wild type. The wings are held at an angle of about 75° from the axis of the body. In this respect it resembles Spread and Dicheate, other third chromosome wing mutants. The surface of the wings may assume any of several conditions. It is usually concave, as though the center portion had grown faster than the wing margin. The two sheets of the wing are frequently in the form of a sack and filled with a clear or brownish liquid. One or both wings of an individual may be affected in this way.

This liquid later may be absorbed leaving the wing in a wrinkled and warped condition. The tip ends of the
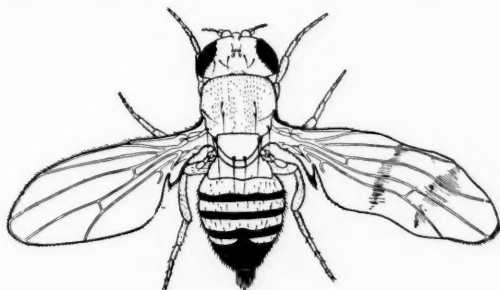

FIG. 1. Taxi wings

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wings are nearly always pinched together or wrinkled. Patches of black pigment are frequently found in various parts of the wings, usually adjacent to a vein or vein intersections.   The venation of the wings appears to be normal.

While the liquid contained within the wings is usually colorless, sometimes it is brownish black, causing the wing sacks to appear black.   Rarely this dark pigment extends through the body, even being noticeable in the eyes.   All the flies of this type which have been observed were females and all died shortly afterward without leaving progeny, so that nothing concerning the hereditary basis has been learned.

Origin of Taxi

On September 30, 1924, there were found, in a stock bottle of apricot-eyed[1] flies, two flies, one female and one male, having a spread type of wings, but the eyes were apricot like those of the culture in which they appeared. These two were isolated but the female had already mated with a normal-winged male, since all but two of her progeny were normal winged.   These normal flies when inbred gave progeny of approximately three normal to one taxi.   The taxi flies mated *inter-se* gave rise to true breeding taxi cultures.

It appeared that taxi arose as the result of a mutation in the apricot culture because all the taxi flies had apricot eyes, which would not have been the case had a contamination from some other spread-wing type of mutant occurred.

The Chromosome of Taxi

Because of its similarity in appearance to curved, a II chromosome recessive, taxi was crossed with it.   The F_1 were all normal and the F_2 gave a 9 + : 3 taxi : 4 curved ratio.   The results, however, were also close enough to 2 + : 1 cu : 1 tx : 0 cu tx, so that a conclusion could not be reached regarding its linkage group from this cross.

[1] An allelomorph of white.

Taxi was then crossed with a Star-Dicheate stock and F₁ Star-Dicheate males back crossed to taxi females, which resulted in the following progeny: 50 S D : 46 D : 43 S tx : 44 tx.

From the association of Star and taxi in the same individuals we inferred that taxi belonged to the III chromosome group of linked genes, and that a 9 : 3 : 4 ratio was obtained when taxi was crossed with curved, the curved-taxi double recessive being phenotypically curved.

Locus of the Gene for Taxi

A preliminary experiment was started to determine the region of the III chromosome containing the taxi gene. A stock containing the recessive spineless, sooty and taxi was established. The F₁ females produced by crossing this stock with wild type were back crossed with spineless-sooty-taxi males. The results of this backcross, given in Table 1, indicate that taxi is near the right end of the chromosome.

TABLE 1

POPULATIONS OBTAINED BY CROSSING FEMALES ss so tx WITH

HOMOZYGOUS MALES $\frac{ss\ so\ tx}{ss\ so\ tx}$

Culture No.	— —		+ —		— +		+ — +		Total
	+	ss so tx	ss	so tx	ss so	tx	so	ss tx	
47 a	92	63	17	14	19	28	7	10	250
47 b	50	33	13	7	14	9	1	7	134
46 a	74	65	17	15	22	21	7	11	232
46 b	62	47	8	9	18	11	1	7	163
Totals	278	208	55	45	73	69	16	35	779
Percentage crossingover			12.8		18.2		4.1 (*)		

Coefficient of coincidence $\dfrac{779 \times 32 \times 100}{174 \times 132} = 108.5$

(*) Calculated on basis of 32 individuals.

The number of flies in the taxi class is lower in each case than the corresponding non-taxi class except in the double crossovers where the ss tx flies are more than

twice as numerous as the double crossover non-taxi class. Other experiments with taxi have shown that taxi flies have a greater accidental death rate than normal winged flies and that in cases where the two classes are expected in equal numbers the normal winged flies regularly exceed the number of taxi flies. Young sooty flies are not readily distinguished from normal body color and it is probable that some ss so tx flies were included in the ss tx double crossover class. In making computations, therefore, from data in Table I, the total number of double crossovers is taken as 32, twice the number in the normal winged class, instead of 51 which is the sum of the two classes as listed in the table which evidently include errors in segregation of classes.

The percentage of coincidence is high (108.5), but this is to be expected because of the long distance involved and the spineless end of this section is in a region of known high crossingover.

The direct crossover value between spineless and sooty, 12.8, checked well with the standard map distance, 12.2, given by Bridges and Morgan.[2]

The crossover percentage, 18.2, between sooty and taxi, indicated that taxi was somewhere in the neighborhood of the gene for rough eyes, which is at 91.1.

In order to secure figures for a more accurate location for the taxi gene, crosses were made to secure a triple recessive stock composed of white-ocelli, 76.2, rough 91.1 and taxi. Several months were spent in the unsuccessful attempt to get recombination between rough and taxi.

This was a strong indication that the gene of taxi was very close to the gene for rough. A triple recessive stock was made up, using ebony, white-ocelli and taxi. The following two of the four possible balanced backcrosses were carried out.

(a) F_1 ♀ $\dfrac{e\ wo\ tx}{}$ $\times$ $\dfrac{e\ wo\ tx}{e\ wo\ tx}$ ♂ and (b) F_1 ♀ $\dfrac{e\ wo}{tx}$ $\times$ $\dfrac{e\ wo\ tx}{e\ wo\ tx}$ ♂.

[2] Bridges, C. B., and T. H. Morgan, 1923. "The third chromosome group of mutant characters of *Drosophila melanogaster*." Carnegie Inst. Publ. No. 327.

TABLE 2

Results Secured by Backcrossing Females e wo tx with $\dfrac{\text{e wo tx}}{\text{e wo tx}}$ Males

Culture no.	e wo tx		+		e		wo tx		e wo		tx		e tx		wo		Totals
	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	
212 A	82	108	142	132	6	2	5	6	20	22	12	23	–	1	1	2	564
212 B	12	9	16	7	–	–	1	–	–	–	3	–	–	–	–	1	48
212 C	77	79	102	108	8	8	6	4	21	15	21	20	–	–	–	1	470
Totals	171	196	260	247	14	10	12	10	41	37	36	43	–	1	1	3	1082

TABLE 3

Results of Backcrossing Females e wo with $\dfrac{\text{e wo tx}}{\text{e wo tx}}$ Males

Culture no.	taxi		e wo		e wo tx		+		e tx		wo		wo tx		e		Totals
	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	
216 A	47	53	43	52	11	11	5	8	5	1	1	3	–	–	–	–	240
216 B	24	30	30	22	5	6	4	5	–	1	1	1	–	–	–	–	129
216 C	5	8	9	15	3	1	2	–	–	1	–	1	–	–	–	–	45
216 D	36	46	49	36	6	7	7	7	3	6	1	1	–	–	1	–	206
Totals	112	137	131	125	25	25	18	20	8	9	3	6	–	–	1	–	620

TABLE 4

Summary of the Figures in Tables 2 and 3, Giving the Percentage of Crossing Over Between the Three Loci, Ebony, White-Ocelli and Taxi

Combinations	—	+—	—+	+—+	Total	Percentage crossing over			
						e wo	wo tx	e tx	Double
e wo tx	874	46	157	5	1082	4.8	15.0	18.8	0.46
e wo tx	505	26	88	1	620	4.3	14.4	18.3	0.16
Totals	1379	72	245	6	1702	4.6	14.8	18.6	0.40

Coefficient of coincidence, 52.1 per cent.

TABLE 5

Cultures from Eggs by Two Females for Successive Three-Day* Intervals in a Ten-Day Period. Numbers Show Percentage of Each Cross-Over Group

Culture no.	—	+—	—+	+—+	Total flies in culture
212 A 1	84.4	2.7	10.1	2.8	109
" A 2	81.0	2.3	16.8	0.0	220
" A 3	82.1	4.8	12.6	0.4	229
212 C 1	78.6	3.4	16.8	1.1	178
" C 2	79.4	3.8	16.8	0.0	238
" C 3	77.0	7.4	15.2	0.4	230

* Parents were removed as soon as larvæ were visible, which on the average was three days.

The summarized data in Table 4, including a total of 1,702 flies, give 4.6 per cent. recombinations between ebony and white-ocelli, which is lower than the standard recombination per cent. of 6.2, as given by Bridges and Morgan, but since both types of backcrosses gave the same approximate results, the variation appears to be a constant one for these cultures. The recombination per-

centage between white-ocelli and taxi is 14.8, which when taken at its face value and using the standard locus of white-ocelli, 76.2, as given by Bridges and Morgan, places the locus of taxi at 91.0, which is only 0.1 of a unit removed from rough. This confirms the indication obtained earlier that the locus of taxi was very close to that for rough. This location of taxi at 91.0 can be considered only as approximate at this time because of the meagerness of the data upon which it is based. With its use in other experiments data should accumulate which can be used to obtain a more precise location.

$$\text{Coincidence} \left(\frac{1702 \times 6 \times 100}{78 \times 251} = 52.1 \right) \quad \text{appears rather}$$

high for this region of the III chromosome where no double crossing over has been found to occur in regions under fifteen units in length. Bridges and Morgan found only three per cent. of coincidence for the Hairless-rough region, which is practically the same as that involved in these cultures with taxi.

General Discussion and Evaluation of Taxi

The ideal arrangement of the genes in determining the locus of a new mutant is to have the unknown flanked on either side by first rank genes, each of which has a definitely established locus, as reference points. In this experiment only one end of the section of the chromosome involved was anchored to a well established locus, that for ebony. The second known locus is not so desirably situated, since it is near to one end of the section, but close enough to taxi, however, that double crossing over in the section between white-ocelli and taxi (14.8 units) can be neglected.

Temperature and age of female are known to effect the rate of crossing-over. The temperature during these experiments was maintained at approximately 24° C. Single females were used for each back cross culture in

each case. The period of egg laying for the females used
was the first ten days after mating. This period was
divided into three or four divisions. The female was
allowed to remain in the first bottle for three days, then
(or when larvæ were apparent) removed to a fresh bottle,
where she remained until larvæ were again visible (about
three days), then transferred to another fresh bottle.
By this method any variations in the crossing over per-
centage, due to increasing age of female, could be ob-
served. However, little variation due to this cause was
expected, for, according to Gowen,[3] the region including
ebony and rough is the least variable portion of the III
chromosome. The results for three successive cultures
from two different females, 212 A and 212 C, are given
in Table 5. No marked differences appear in these
cultures.

Interesting relations have been found to exist between
taxi and other genes affecting wing characters. It appears
that the gene for taxi can produce its characteristic effect
only when all other genes affecting wing development are
of the normal type, that is, the taxi gene interacting with
normal wing genes produces taxi wings. If one of the
many genes which result in the production of normal
wings is altered by mutation the taxi gene is rendered
ineffective in development.

The monohybrid recessive vestigial when crossed with
taxi produces a normal F_1 type and $9 +$: 3 tx : 4 vg in
F_2. The double recessive, vgvgtxtx, is vestigial in ap-
pearance and when bred with pure vestigial vgvgTxTx
produces only vestigial offspring. When crossed with
taxi, VgVgtxtx, it produces taxi in F_1 and 3 tx : 1 vg
in F_2.

Taxi shows similar relations with two other monohy-
brid recessive II chromosome wing mutants, clip (T^{cp})
and curved (c).

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[3] Gowen, J. W., 1919. ''A biometrical study of crossing over. On the
mechanism of crossing over in the third chromosome of *Drosophila melano-
gaster.*'' *Genetics* 4: 205–250.

The old unit character idea which implied that a given character was the result of the presence or absence of a single gene in the germinal material has been replaced by the theory that a given character is the result of the interaction or cooperation of many genes, one of which may act as a differentiator for a given expression of that character. Thus the fact that many mutant wing types in Drosophila each show allelomorphic relations with the wild type but are known not to be members of a multiple allelomorphic series, establishes the multiple gene basis for the development of the normal wing type. A mutation in any one of these numerous loci alters the system to the extent that the end product of the interaction of the series of multiple genes controlling wing development is different and then a ''mutant'' character appears. The mutant gene now acts as a differentiator of an interacting system of genes, all of which are necessary for the production of the character.

The hypostatic nature of taxi, however, shows that some of the II chromosome genes for wing characters are able to produce their typical expression irrespective of the type of gene (tx or Tx) present in one locus of the III chromosome, and conversely that taxi wings develop only when the taxi gene interacts with the normal allelomorph of the vestigial, curved and clip loci.

Taxi flies have a lower survival value than wild type. In Table 1, considering the non-taxi flies as 100 per cent. in viability, the taxi flies show 85 per cent. viability. This is frequently true of recessive mutants. The Mendelian proportions, however, are not seriously altered by the lower viability of taxi.

Taxi is easily and readily separated from the normal and has been found an excellent III chromosome character for use in laboratory instruction. It makes a good alternative character for rough and has the advantage that magnification is not necessary for its recognition.

Summary

1. Taxi wings is a new III chromosome monohybrid recessive similar in appearance to spread and to Dicheate.

2. Its viability is slightly lower than the viability of comparable, normal-winged flies.

3. Taxi is hypostatic to vestigial, curved and clip, three II chromosome recessives.

4. The locus of the gene for taxi is approximately 91.0, very near to the locus of the gene for rough eyes.

5. Taxi supplies a good marker for the right end of the III chromosome and can be used as an alternate for rough.

SOME HYPOTHESES CONCERNING THE PHYLOGENY OF THE ALGAE

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EVERY one who has observed and considered a field of marine algae on the seashore at low tide knows that there are four very conspicuous groups of plants in the ocean. They are the blue-green, red, brown and green algae. The obvious question is, Why does the sea contain plants of such a variety of colors when the land supports only green vegetation? How is one to dispose of the problem of the origin of the different phyla of marine plants?

In the algae, morphological and taxonomic evidence does not satisfactorily explain relationships in the form of a phylogenetic tree. Conclusions arising from such evidence make necessary the consideration of other criteria of evolution. The writer is convinced that the theory of parallel development contains the solution of the problem. However, in connection with parallel development, it is necessary to bring forward an explanation having reference to physical conditions prevailing through long periods of time during the early history of living forms.

The following brief outline is offered as an introduction to this paper:

1. A parallel development of types, rather than the usually accepted tree-like form, is to be regarded as the course followed in the evolution of the different phyla of marine algae.

2. The four principal groups of marine algae are considered to be radically different from each other, and especially in so far as pigmentation is concerned. The special pigmentation character, belonging to each, must be acknowledged to be a heritable character of high permanence, for it evidently was so impressed upon the

organism during its early history that certain types of
the four races of algae are to-day found growing side by
side under identical environments.

3. It is here suggested that the various pigments in-
volved were originally called into existence in response
to certain physical environmental forces or variables and
that by far the most important of these external factors
was illumination, the quantity or quality of which was
determined by or modified by two different media (a),
the sea water in which the organisms lived and (b), the
layer of atmosphere above it.

4. The present day distribution of marine algae is in
agreement with these theories. The Rhodophyceae and
the Phaeophyceae—for example, the large kelps—
flourish under weak illumination, as in the temperate
zones wherever fogs and gloomy skies prevail during the
greater part of the year, and throughout the polar re-
gions, where such plants are found reproducing freely
during the long dark winter nights. On the other hand,
the Chlorophyceae reach their most luxuriant develop-
ment under the brilliant sunlight of the tropics. The
Myxophyceae, or blue-green algae, apparently are found
everywhere. The fact that most of these plants have
thick individual gelatinous sheaths and that in addition
they almost invariably occur in colonies of their own kind
or among masses of other floating algae, is enough to
prove that they perhaps, more than any of the other
types, are protected from intense light, as far as the indi-
vidual protoplast is concerned.

It is impossible, in the opinion of the writer, to inter-
pret the present distribution of marine algae without
taking under consideration the changes which have oc-
curred during the long geological periods of time involved
in the history of the world and the effect of these changes
upon life existent in its waters.

It may be taken for granted that the earth during its
infancy or early childhood was a core of rock material,
the "lithosphere," surrounded by a shell of air, the
"atmosphere," and that later there appeared between

them a third—discontinuous—shell of water, a gradual accumulation through volcanoes and thermal springs, the "hydrosphere." Here we have in mind Dr. Chamberlin's picture of the juvenile planet, "a small lithosphere, a small atmosphere, and a small hydrosphere growing up together in cooperation and at the same time in competition and antagonism, and so, by their interactions, working out their adjustments to one another progressively."[1] Whatever may have been the initial history of the earth, it is safe to assume that the water, as it was slowly liberated by the earth itself, was at first at least somewhat warm or even hot. In connection with this paper the important point to consider is, however, that the physical condition and chemical composition of both water and air layers must have undergone changes as time went on. The entire atmospheric shell was probably initially very much denser than in later eras. From complete darkness, through every grade of grayness, to the bright white light of to-day, is the history of *illumination* so far as it had to do with life on this planet. Not for long ages, perhaps, did the sun burst in its full intensity upon the surface of the waters. Whether the density of the atmosphere was due to the plunge of planetesimals into the upper layers of air, or to the presence of much water vapor, or to some chemical condition, matters little for the purpose of the present paper.

For convenience of discussion, merely, geological time is divided into "Epochs" and "Ages" ("Eras") as indicated diagrammatically in the accompanying figure (Fig. 1), but these arbitrary time divisions have not been definitely related to geological divisions. The author believes that most of them precede the oldest geological divisions as determined by paleontological evidence. The "Age of Chlorophyceae" probably falls near the beginning of the geological time scale.

Life probably appeared in the waters of the earth much earlier than we are accustomed to believe. It is sufficient

[1] Chamberlin, T. C. The origin of the earth. Chicago, 1916, p. 168.
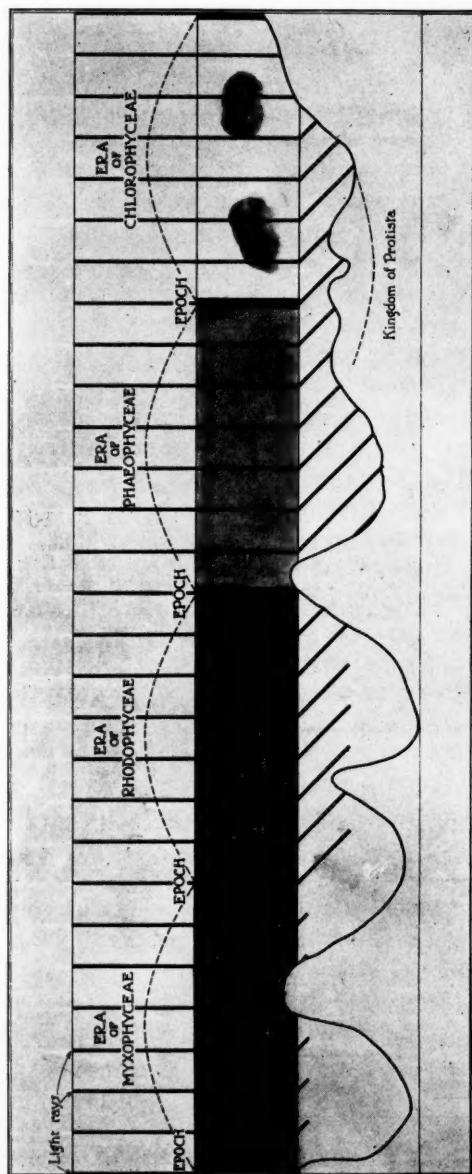
FIG. 1. Diagrammatic representation of division of geological time into phycological "Epochs" and "Ages" or "Eras"

to say that it appeared when conditions on this planet were ready for it—initiated, it may be, in the form of morphologically structureless masses of protoplasm, without limiting wall and without nuclei.   Whatever theory one wishes to adopt concerning evolution, all must agree that the primitive organism must have possessed at least three capacities: the ability to make its own food; the ability to reproduce itself; the ability to respond to the environment.

Judging from such knowledge as we possess to-day with regard to plant life, *viz.*, that light and chlorophyll are necessary for the manufacture of food, we can take for granted two statements:

1. Some form of chlorophyll, or related pigment, constructed from materials at hand in the sea water, must have formed a part of the body of the primitive organism.
2. Light from the sun must have penetrated through the atmospheric shell, at least to a certain extent.

It must now be asked if there are existing types which might indicate ancestors corresponding to such primitive life forms as those suggested.   The answer is found in the group of the blue-green algae, or Myxophyceae, which contains just such organisms.   Here are one-celled plants and extremely simple multicellular plants.   We, therefore, seem justified in designating the first "Era" ("Age") shown on the chart the "Age of Blue-green Algae."

1. Myxophyceae.   Blue-green Algae.   The so-called "cell" of the blue-green alga consists of a mass of protoplasm surrounded by an extremely thin membrane which, as recent studies indicate, is in reality a modified outer layer of protoplasm rather than a true cell wall.   After a complete re-survey of the subject Dr. S. Prát[2] concludes

[2] Prát, S.   "Beitrage zur Kenntnis der Organization der Cyanophyceen." *Archiv für Protistenkunde,* 1925, Vol. 52, pp. 142–165.   1 plate and three text-figures.

that no nucleus is present in these organisms. The blue-green organism, then, is a mass of protoplasm without limiting wall and without nucleus.

The protoplast seems to consist of a central mass of colorless cytoplasm surrounded by a layer of colored cytoplasm, usually of a characteristic blue-green tint, but in some cases olive-green, rose or violet. These hues are due to the presence of primarily three, or rarely four, pigments, not here associated into chromatophores as in the higher plants, but of so rudimentary a structure that no student has yet been able to properly describe them or to announce the formulae for their chemical composition.

The most abundant of these pigments is *phycocyanin,* a water soluble protein substance, blue in color, apparently existing in a diffuse state in the cell.

Chlorophyll, the second pigment, seems to be present in the cytoplasm in the form of very minute aggregations and is more or less hidden or masked by the phycocyanin, the two together giving the "blue-green" color characteristic of the Myxophyceae.

The third pigment, *carotin,* is orange in color and it likewise apparently occurs in a granular form. In structure both chlorophyll and carotin seem to be different from the forms of the same pigments associated in the chloroplastid of the higher plants.

In rare cases, as in certain species of *Oscillatoria* and *Phormidium,* a fourth pigment, *phycoerythrin,* is present. This is the characteristic pigment of the red algae and will be discussed later.

It is important to note that true starch is not found in these rudimentary plants. The storage material in the blue-green algae is *glycogen.*

The blue-green algae reproduce themselves in the simplest possible manner. The unicellular individual merely divides itself into two daughter plants. Multicellular plants break up into short chains of cells, each one capable of developing into a new plant. A primitive type of resting cell is formed in some groups.

In conformity with the theory that the blue-green algae now existing have arisen from the first pioneers on the frontiers of life itself, and that the ancestral forms came into being during the period of weakest illumination, the first era of life on this globe, then it must be acknowledged that those ancestors passed through every possible hardship in the world's history. This is indicated by the fact that the species alive to-day are still able to endure practically any adverse conditions which may confront them. They are found on all parts of the globe, in widely differing conditions of heat and cold, of dryness and moisture, of shade and sunlight. They flourish in salt water and in fresh, in cold water and in thermal springs. They are everywhere. Here may lie the explanation of the presence of blue-green algae in the waters of hot springs. We know that the strictly thermal algae are all low forms of the Myxophyceae, and also that the same species, as, for example, *Phormidium laminosum,* are found in practically all hot spring localities, in Yellowstone Park, in California, at Rotorua and Whakarewarewa, New Zealand, and elsewhere. These plants must have been universally distributed in the first warm waters of the young earth. Their descendants are the hot spring algae of to-day, for it is not impossible to suppose that they may have been kept in existence by means of aerial distribution, as are their near relatives, the Bacteria. Many of them secrete lime or silica in connection with their life processes, and desiccated particles of silicious sinter and of travertine, blowing about as dust, carrying with them minute bits of gelatine containing live plants or resting cells, from the drying rims of thermal springs and geysers, would serve to start new growths wherever they might come in contact with thermal conditions.

It is interesting here to note that in Minnesota lakes blue-green algae form "water-blooms" during the hottest days of summer. Quoting from Dr. G. M. Smith,[3]

[3] Smith, G. M. "Ecology of the Plankton Algae in the Palisades Interstate Park," 1924, p. 110.

"Broadly speaking, the blue-green algae thrive best at summer temperatures." He goes on to say that Wesenberg-Lund[4] "maintains that temperature plays an important rôle in 'blooming,' and that those species that require warm midsummer temperatures of the water for maximum growth do not appear in quantity in cool summers when the temperature of the water does not rise above 18 degrees C."

Now, Dr. Smith, in the paper cited above, speaking of blue-green algae, also notes that, "although light is essential for the growth of algae, very intense light may *not* be favorable. . . . This direct killing effect of the sun's rays may be of much greater importance than has been suspected, and may account for the fact that the surface of a lake frequently contains fewer algae than the region just below the surface." This effect of the intensity of the sun's rays also has been noted by Fritsch[5] in a discussion of the algal flora of Ceylon. He suggests that the "great predominance of the blue-green algae in the tropics may be due to a protection of the green coloring matter from the intense light by the blue-coloring matter."

So the question will be asked: "If the blue-green algae came into existence during the period of weakest illumination, how can one account for the fact that they seem to be very well able to endure the bright light of the present time?" It is easy to answer this question by pointing to the fact that every blue-green plant is either fully protected from light by an individual gelatinous sheath, or lives in common with its associates within a relatively large mass of gelatinous matter secreted by all the individuals in the colony. A few unicellular plants do not possess a sheath and do not occur as colonial forms. These, however, will always be found living under pro-

4 Wesenberg-Lund, C. "Studier over de Danske soers plankton." Kjobenhavn, 1904.

5 Fritsch, F. E. "A General Consideration of the Subaerial and Freshwater Algal Flora of Ceylon." A contribution to the study of tropical algal ecology. I. Subaerial Algae and Algae of the Inland Fresh Waters. *Proc. Roy. Soc.* London, Ser. B, 1907, Vol. 79, pp. 197–254.

tection from light in masses of other algae, as the green algae which form layers and mats on the surface of ponds.

The writer believes that under present conditions the blue-green algae have made use of their gelatinous sheath material to shield themselves from too strong illumination and that their distribution to-day is largely due to temperature.

The bacteria appear to be closely related to the blue-green algae and may have originated from them, or near them. Both groups contain certain forms which are able to maintain life under the highest temperatures known for living organisms. In both groups the protoplast is without a nucleus. Possibly under the severe test of some land conditions certain forms of the blue-green algae learned to live on organic matter and eventually lost their chlorophyll and other pigments. It has long been the opinion of the writer that certain sea-forms of the blue-green algae are now far on the way to becoming at least partial parasites and saprophytes. Toward the end of the summer season the battered apices of fronds of the large red and brown algae may be observed to be covered by a growth of blue-green forms. One is strongly reminded of similar growths of bacteria and fungi on weakened portions of land plants.

2. Rhodophyceae. Red Algae. The second "epoch" must have been one concerned with the lessening of the density of the atmosphere. Stronger rays of the sun, penetrating through the thinner screen and through the layer of sea water, according to the laws of physics, of chemistry and of life, would call into being in the protoplasmic contents of the organism a pigment of a different structure. *Phycoerythrin* is a pigment which would correspond to such needs. It is the principal pigment of the red algae, Rhodophyceae, and masks the other pigments contained in the chromatophore—a new structure in the plant cell. According to Palmer,[6] phycoerythrin is not a

[6] Palmer, L. S.   Carotinoids and related pigments.   New York, 1922, pp. 100–102.

carotinoid. It is a colloidal, nitrogenous substance allied
to the proteins, soluble in water, but not in organic sol-
vents. Purified solutions of phycoerythrin are very
difficult to secure, and apparently no satisfactory analy-
sis to indicate its composition has yet been obtained.

Considerable study has been made on the amount of
light penetration into the water. In deep water blue-
green light preponderates which chlorophyll alone could
not adequately absorb; therefore it is probable that phy-
coerythrin provides effective light absorption in such
regions. Atkins and Poole,[7] in a recent paper, state that
the absorption due to the water, as such, results in the
longer wave-lengths being reduced in far greater propor-
tion than are the blue and violet; that, indeed, the ultra-
violet transmission may be surprisingly large in very
clear water. Quoting these authors, ''The red end of the
spectrum, then, being cut out, the light remaining is
largely blue in clear water, so red algae are obviously
efficient absorbers of light; they flourish, therefore, in
clear water, where their red pigment is an adaptation that
enables them to utilize to advantage the blue light present
in reduced amount.'' In the light of the present theory,
however, it may be suggested that the red algae flourish
in clear water where the blue light is present in reduced
amount because under just such conditions of illumina-
tion their ancestors were developed. Miss K. M. Good-
win,[8] in a paper published in March, 1926, makes the very
interesting statement, ''Phycoerythrin assists assimila-
tion by absorbing blue-green light and degrading it to
those wave-lengths absorbed by chlorophyll.'' So little
is known to-day of the rôle that phycoerythrin plays in
the economy of the red algae.

But the chromatophore of the red algae contains other
pigments: chlorophyll, carotin, xanthophyll—and in rare
cases phycocyanin—is present. Since it is now known
that a few blue-green algae are provided with phyco-

[7] Poole, H. H., and Atkins, W. R. G. *Jour. Marine Biol. Assoc.* n. s.,
1926, Vol. 14, pp. 177–198.

[8] Goodwin, K. M. *New Phytologist*, 1926, Vol. 25, pp. 51–54.

erythrin and that some red algae contain phycocyanin, we have evidence that certain ancestors of existing red algae must have arisen side by side with early forms of certain blue-green algae.

It should be noted here that, according to Dr. F. Schütt,[9] phycoerythrin has an intensive orange-yellow fluorescence.

With chlorophyll present in a chromatophore, accompanied by at least carotin and xanthophyll, it is very interesting to note that the product of metabolic activity is not true starch, but what is termed "Floridean starch." Nor is this substance closely connected with the chromatophore, at least in all cases. The comparatively simple red alga, *Nemalion,* is said to possess true pyrenoids which are actively concerned in the elaboration of the soluble Floridean starch, lying diffused throughout the cytoplasm of the cell. A still more simple red alga, *Porphyra,* possesses a high type of pyrenoid, but in most of the genera of the group this body is entirely absent.

It is here suggested that the "pyrenoid" perhaps came into existence in response to the strong light conditions of shallow shore waters. The only marine algae possessing pyrenoids appear to be the green forms in general, and a limited number of brown and red algae, as *Ectocarpus,* a genus of the Phaeophyceae, and *Porphyra* and *Nemalion,* members of the Rhodophyceae. The green forms which succeeded in maintaining life in the fresh waters of the land in almost all cases have pyrenoids. One of the rare instances in which pyrenoids are found in forms above the algae is the liverwort, *Anthoceros.* These facts would lead one to believe that the few red and brown algae which possess pyrenoids are of much later origin than those which are without them; in other words, that the Eu-Florideae comprise the ancient forms of the Red Algae, while the Bangioideae (the Protoflorideae of Rosenvinge) constitute the newest forms which have appeared in the ocean.

[9] Schütt, F. *Ber deutsch. Bot. Gesellsch.*, 1888, Vol. 6, pp. 36–51; 305–323.

From simple one-celled plants the elaborately con-
structed red seaweeds of the present day have gradually
evolved, supposedly in response to various factors of
temperature, composition of water, currents, tides and
wave action, and shore features. But, while the sexual
reproductive system of the higher red algae is almost
equal to that of the flowering plants in general complex-
ity, there is no indication in any known species that a
ciliated cell has ever been a part of the reproductive sys-
tem of the plant.

3. Phaeophyceae. Brown Algae. In the third "era"
(age) the penetration of the radiance of the sun attained
fuller power, but by no means equalled that of the present
time. The rays of light passing through the thinner layer
of screening material in the atmosphere were able to
penetrate much more deeply into the water itself. Shal-
low seas with wide expanses of shelving shores and ex-
tensive tide-pool formations were in existence at this
time and new forms of life, ancestors of the brown algae,
could appear in response to the new conditions of illumi-
nation characterizing the "third age" or "era."

Shallow water and stronger illumination favored the
appearance and persistence of a new type of organism
which was characterized by motility, and with few excep-
tions the brown algae of the present time possess ciliated
reproductive bodies.

The special feature of interest in the motile cell of the
brown algae is the point of insertion of the cilia. The
pyriform body resembles the ordinary type of "swarm-
spore" in a general way, but the pair of cilia arises at
one side of the pointed anterior end of the zoid. The
cilia are unequal in length and, while the longer one
points forward, the shorter one trails in the rear. Dr.
Church[10] is probably correct in assuming that this feature
—the insertion of the cilia—was originally an adaptation
to light. The organism, when it sank too deeply in the
water, was fitted to move upwards with a spiral swim-

[10] Church, A. H. *Oxford Botanical Memoirs,* 1919.

ming motion until it reached the position of its optimum illumination, but this spiral climbing motion is not adapted to precise movements. The lateral insertion of the cilia is a feature of the brown algae to-day.

The one fundamental character, apparently, which has remained unchanged throughout all time in these products of the "Age of Phaeophyceae," or the third era of illumination, is a new pigment, *fucoxanthin,* brown in color, which must have been needed to absorb certain light rays in connection with the chlorophyll-system.

Recently a careful study has been made of the pigments which constitute the chromatophore of the brown algae. Miss Howard[11] has proved the presence of five separate pigments: Chlorophyll *a* (pale green); chlorophyll *b* (deeper green); carotin (yellow or orange); xanthophyll (very pale yellow); fucoxanthin (yellow or brown). Living plants, the author points out, owe their brown tint to the high concentration of the carotinoid, *fucoxanthin.* (Dead plants owe their color to the pigment phycophain, a post-mortem product.)

Willstätter's formulae for the five pigments:

Chlorophyll *a*	$C_{55}H_{72}O_5N_4Mg$
Chlorophyll *b*	$C_{55}H_{70}O_6N_4Mg$
Carotin	$C_{40}H_{56}$
Xanthophyll	$C_{40}H_{56}O_2$
Fucoxanthin	$C_{40}H_{56}O_6$ (or $C_{40}H_{54}O_6$)

Two interesting quotations from recent works are given in connection with this matter. The first one is from Sharp's "Introduction to Cytology":[12] "With chlorophyll are usually associated one or both of the yellow carotinoid pigments, carotin and xanthophyll, which seem to be similar to the lipochromes of animals. There also appears to be a close chemical relationship between chlorophyll and the haemoglobin of animal blood. The exact mutual relation of the several pigments in the

[11] Howard, Grace E. Extraction and separation of the pigments of Nereocystis luetkeana. Publ. Puget Sound Biol. Station, 1921, Vol. 3, pp. 79–91.

[12] Sharp, L. W. "An Introduction to Cytology." New York, 1926, p. 100.

chloroplast is uncertain, some investigators thinking it probable that a single green compound decomposes readily into 'chlorophyll *a*,' 'chlorophyll *b*,' and the yellow pigments when subjected to analysis.''

Willstätter and Stoll[13] make the following suggestion: ''Carbon dioxide, attracted to the chlorophyll system by virtue of the magnesium it contains, is reduced by 'chlorophyll *a*' (by the agency of light energy), which itself becomes 'chlorophyll *b*.' As we saw, 'chlorophyll *b*' is an oxidation product of 'chlorophyll *a*,' containing one atom more oxygen. Two molecules of 'chlorophyll *b*' might then give off a molecule of oxygen, and become 'chlorophyll *a*' again. It is further suggested that this removal of oxygen may be the function of carotin, which thereby becomes xanthophyll. A reducing enzyme finally converts xanthophyll into carotin again. But this naturally is, at present, purely hypothetical.''

The assimilation product of the brown algae is *pentosan* $(C_5H_8O_4)n$, which resembles the true carbohydrates in its chemical composition and properties.

There is evidence that during the latter portion of the ''Age of Phaeophyceae'' a large number of different types arose, all more or less resembling the unicellular type of the Brown Algae. They all possessed nuclei. Most of them were motile, either by means of two lateral, unequal cilia, one flagellum, or protoplasmic currents. In relation to the present problem their most important characteristic is their pigmentation. Those of their descendants which we know to-day agree in the possession of golden brown chromatophores, some exhibiting in addition a pyrenoid and some an eye-spot or pigment-spot. The assimilation product appears to be in most cases a fatty oil, but a beginning is made, on the part of some, to store starchy material as a product of photosynthetic activity. Some of these types are now animal-like in their method of taking food, ingesting solid particles, and some of them are saprophytic. The writer refers here to the

[13] Willstätter, R., and Stoll, A. ''Untersuchungen über Chlorophyll.'' Berlin, 1913, p. 25.

groups of Flagellates, Dinoflagellates (Peridiniales), the unicellular members of the Heterokonteae and the Diatoms. It is thought that the ancestors of these groups had their beginning within close propinquity to the brown algae and that they were the earliest members of the "Kingdom of Protista." The algae of the group Heterokonteae owe their brownish color to an excess of xanthophyll in the chromatophore.

It may be well to review very briefly a small part of the literature dealing with the algal pigments, phycocyanin, phycoerythrin and fucoxanthin, before passing on to the green algae.

Englemann[14] in 1884 first stated the hypothesis that the color of marine algae was complementary to the color of the light to which they were exposed. He considered that the production of pigments was the reaction to a particular factor in the environment, the frequency of the light in which the plants grow.

Richter (1912),[15] on the other hand, contended that it is the intensity of the light which determines the color of the algae, and that the distribution of the different colored algae is a matter of light intensity entirely analogous to "light" and "shade" land plants.

Shelford and Gail,[16] working in the Puget Sound region, give the depth for the red algal zone from ten to thirty meters; the depth for the brown algal zone as from five to twenty meters. In the red algal zone the blue light is 2 to 10 per cent. of full sunlight and the red light is 0.032 to 1 per cent. In the brown algal zone the shorter wave-lengths have an intensity of about 10 per cent. of full sunlight and the longer red wave-lengths about 1 per cent.

In 1921 Dr. Francis E. Lloyd[17] stated that, "in general the blue-green species which contain phycocyanin are

[14] Englemann, T. W.   *Botanische Zeitung*, 1884, Vol. 42, p. 81.

[15] Richter, P.   *Berichte Deutsch. Bot. Gesellsch.*, 1912, Vol. 30, p. 280.

[16] Shelford, V. E., and Gail, F. W.   Publ. Puget Sound Biol. Station, 1920, p. 141.

[17] Lloyd, F. E.   *Science*, 1923, Vol. 58, p. 91 and 229; 1924, Vol. 59, p. 241.

fluorescent red, while the brownish or violet tinged kinds
containing phycoerythrin are orange fluorescent.'' Pro-
fessor Lloyd thinks that the fluorescent pigments involved
may all prove to be of much greater importance than at
present supposed (chlorophyll excepted). He suggests
the possible significance of the physiological studies in
the realm of photodynamics and their bearing on the
nature of the chlorophyll mechanism, and he is sure that
further extended and more critical study of the water-
soluble fluorescent pigments will bring a rich victory.
''One can not be aware of the discussions in the field of
photo-therapy, as the remarkable curative effect of light
in rickets, without feeling that there exists some relation
between this and the fluorescence of the blood pigment.''

Dr. H. A. Spoehr[18] has recently reviewed the subject
of algal pigments but adds no new information. ''The
red pigment, phycoerythrin, and the blue pigment, phy-
cocyanin, though extensively investigated, have as yet not
been subjected to exhaustive chemical study.   They are
complex nitrogenous substances, related to protein but
of a lower nitrogen content.   There has been much specu-
lation regarding the possible rôle which these pigments
play in the photosynthetic process, though no satisfactory
conclusions have been arrived at.   Fucoxanthin is a
carotinoid and its absorption spectrum is similar to that
of carotin and xanthophyll.   Phycoerythrin and phy-
cocyanin are strongly fluorescent, a property which has
been made the subject of much study.''

The fact that so much work has been done with such
meager results brings out strongly the need for the physi-
cist in this field of botany (and medicine). Dr. Spoehr
draws attention to this point.   ''A perusal of the many
investigations undertaken in the field of photosynthesis
leads to the conclusion that the cause for the disappoint-
ing outcome of many of these elaborate and costly experi-
ments lies in a disregard of some of the fundamental prin-
ciples of the physics of light.   There is a wealth of

[18] Spoehr, H. A. ''Photosynthesis,'' New York, 1926, p. 345.

information originating in physical and chemical laboratories which has direct application to these problems and can not be disregarded if intelligible results are to be hoped for.''

Before leaving the division dealing with the brown algae, attention should be called to the fact that they are the forms which to-day have a definite capacity for storing iodine from sea-water. This might suggest a difference in the chemical composition of the sea-water in which these organisms originally lived.

4. Chlorophyceae. Green Algae. At the beginning of the ''fourth age'' or ''era,'' either the atmosphere lost considerable of its water vapor which broke up into the floating masses we call ''clouds,'' or there was some change in the chemical composition of the atmosphere, or it became freed of planetesimal dust for the first time. The direct rays of the sun in its full intensity fell upon the surface of the water and the broad shelving coasts.

Such new conditions would strongly affect life in the waters. New kinds of organisms must have come into being, plants which were able to evolve true, morphologically differentiated chloroplastids as we know them now, and the pyrenoid became an essential part of the chloroplastid. For the first time in the history of plant life true starch was formed as the product of metabolic activity. Evidently accessory pigments were no longer needed as the chloroplastid containing ''chlorophyll *a*,'' ''chlorophyll *b*,'' carotin and xanthophyll, was exactly fitted to respond to the direct rays of the sun which no longer filtered through an atmospheric layer obstructive to sunlight.

Great numbers of new forms must have arisen in response to the changed order of things. The vast shallow seas, teeming with life, were, in truth, the very ''cradle'' of the Kingdom of Protista. Unicellular brown algae, unicellular green algae, unicellular animal forms developing from, or side by side with, unicellular algae, must have existed in countless multitudes. An intensive study

of one-celled algae must lead one to agree with Haeckel that the polyphyletic origin of the analytic protozoa from the synthetic protophyta is unmistakable.

At the present time almost all species of green algae possess, at least at some phase of the life cycle, ciliated reproductive cells, but the swimming apparatus of the green cell is in almost all cases different from that of the brown algae. In them, a pair (sometimes four, or even a crown or circlet) of cilia of equal length is developed from the anterior end of the body. Perhaps this allows the motile cell to move directly toward a definite point and it may be that these changes in its form were an adaptation to a lateral mode of progression induced by the proximity to land and the low tide conditions afforded by the presence of numerous small tide-pools.

During the "Age of Chlorophyceae" must have begun the migration to the land, on the part of certain kinds of green algae. Space and food materials along the shores of continents and islands were limited. The struggle for existence must have led to the expansion from tidal conditions to freshwater and aerial conditions. There was the innate, inevitable tendency to cover, or to occupy, all available space—the end of which was to be the clothing of every continent and island with green plants. Because of the barrier of brilliant illumination neither brown nor red algae could ever adapt themselves to land conditions, unless, as some think, the latter developed into the higher groups of Fungi through a change in food economy and a consequent loss of pigments. The blue-green algae, because of their ability to protect their protoplasts from the too strong light by means of secreted gelatinous material in the form of sheaths, were able to establish themselves upon the land and to exist there under the most adverse circumstances, but the very adaptive character which made them able to occupy dry land precluded their further evolution with the result that they still remain very low in the scale of life. In many of the higher Myxophyceae resting cells have been evolved and there

are a few cases of true branching.   Beyond that these
plants have made little or no progress.

It may be said that of all the forms which left the in-
tense competition of the shore waters for the hitherto un-
occupied terrain, the only true successes, in the fullest
sense of the term, were the green algae.   In themselves
they are not high types of plant life, but one can not
doubt that their ancestors and the ancestors of the vari-
ous phyla of existing land plants were closely related.

As a last word, let us consider for a moment those
primitive bits of life matter representing the ancestral
forms of the four principal algal groups.   Given to each
its little packet of chlorophyll with which to make its
food, what must we credit them as actually having accom-
plished in the way of "responding to the environment"?
Certainly, the production, in each particular case, of a
distinct and characteristic pigment, nicely adapted to cer-
tain ranges of illumination, was a fundamental attain-
ment.   It may be that the relation between the specific
amount of illumination and the character of the pigment
needed to receive the penetrating rays in order to make
them available for the use of the chlorophyll was the
cause of the beautifully colored seaweeds on our shores
to-day, and the reason why the plants of the land are
green and not *red* or *brown.*

The author desires to express her appreciation to those
of her associates who have read the above paper in manu-
script and have been kind enough to criticize it.

TOPICS FROM THE LIFE HISTORY AND HABITS OF THE RED–BACKED SALAMANDER IN SOUTHERN MICHIGAN[1]

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SEVERAL aspects of the life history and habits of the red-backed salamander, *Plethodon cinereus cinereus* (Green), that are in need of new or further data have been studied by the writer in the vicinity of Ann Arbor, Michigan, and the data obtained summarized here as a contribution to a fuller understanding of this much-studied but still incompletely understood species. Field work has been limited to chance opportunities during the autumn and spring months from September to May (but largely exclusive of these months) in the years 1923 to 1927. Some of the topics taken up have apparently not been treated before for this species, namely, the mating season, spermatophores and the breeding age; others, sex differences, color phases, egg complements and food habits, have been discussed at more or less length by earlier writers.

The most extensive studies of this species are those of Piersol (1910 and 1915) and Cochran (1911). Dunn has summarized all previous work in his "Salamanders of the Family Plethodontidae" (1926) and here will be found a full bibliography. A list of some of the more useful references bearing on the life history and habits of this species has been put at the end of this paper.

Sex differences.—That sexually mature males differ from sexually mature females in the swollen naso-labial glands and the elongated premaxillary teeth has already been pointed out by Dunn (1926) and by Noble (1927). A difference in length of body is brought out by measure-

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[1] Contribution from the Zoölogical Laboratory of the University of Michigan.

ments on a series of 124 males and 101 females. The mea-
surements were taken from the tip of the snout to the
posterior part of the hind leg at its junction with the
body. The males measured from 35 to 47 millimeters and
the females from 38 to 48 millimeters. A plotting of all
measurements (Fig. 1) shows that males, as a rule, are
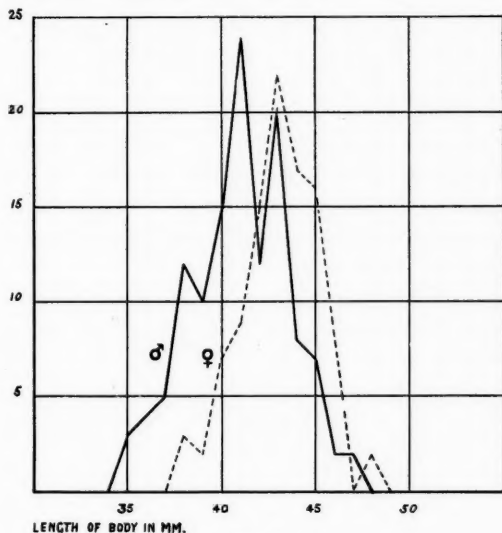shorter in body length than females.

LENGTH OF BODY IN MM.

Fig. 1

Color phases.—The red-backed and black-backed phases
varied in relative numbers in every collection, but the
author has searched in vain for evidence that the indi-
viduals of the two phases are otherwise than equally

Locality in vicinity of Ann Arbor	Males		Females	
	Red	Black	Red	Black
Iosco	56	56	33	48
Freedom	14	18	15	8
Pittsfield	13	9	17	9
Other places	16	9	16	11
Totals	99	92	81	76

numerous in any locality and between the sexes. The accompanying table shows the variation found in three collecting places, and in the lowermost row is given a summary of all specimens tabulated by the writer for the vicinity of Ann Arbor, Michigan.

It will be noted that when the summary is completed by color there are found to be 180 red individuals and 168 black ones. This is 51.7 per cent. red-backed and 48.3 per cent. black-backed, which is practically a ratio of one to one.
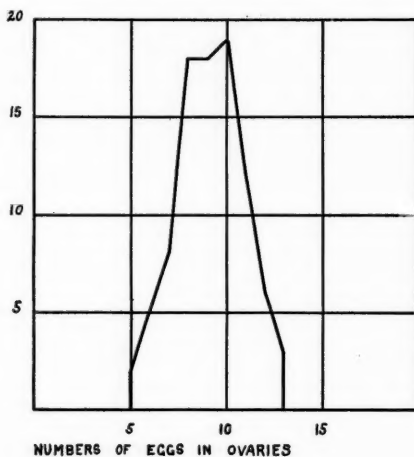

NUMBERS OF EGGS IN OVARIES
Fig. 2

Egg complements.—Dissection of 91 mature females showed a variation in total egg complements of 5 to 13 (exclusive of the under-sized eggs that are frequently present and that are referred to below). The results are shown in the diagram of the number of eggs (Fig. 2). Of the eggs in these counts, 423 were in left ovaries and 402 in right ovaries, or, in per cent., 51.3 per cent. of these eggs were in left ovaries and 48.7 per cent. in right. It is very common to find, in one ovary or the other, two or even three under-sized eggs, the reason for which is not clear. Piersol commented on the presence in the egg clusters of occasional small eggs. It is possible that

these small eggs, like the eggs from the interior of the cluster, give rise to weak and small individuals. This should be checked by observation. Perhaps these small eggs are not all laid.

There appears to be no correspondence between the size of the female and the total number of eggs produced, —that is, small females will deposit as many eggs as large females. This statement is based on the figures in the accompanying diagram, which shows the relation between length of body and number of eggs produced (Fig. 3).

Breeding age.—One hundred and ten individuals collected October 31, 1926, fell into three groups on the basis
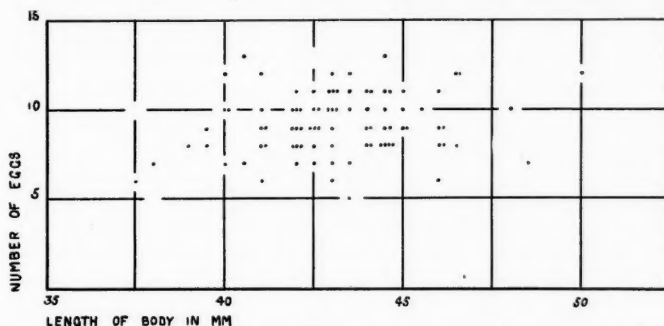

FIG. 3

of size. The group of smallest individuals was plainly composed of the young of the year and the group of largest ones was almost exclusively of sexually mature individuals. These groups stand out clearly when the figures are plotted. If to the individuals of this collection be added those taken on October 24 and 29 and November 5 and 9 in 1924, the three size groups are still more plainly evident (Fig. 4). From these figures it is assumed that individuals of this species ordinarily attain sexual maturity at the end of their third season, or in a little more than, or about, two years after hatching, and that eggs are laid in the fourth season, *i.e.,* when the individuals are less than three years old. The few specimens intermediate between the groups may be precocious or slow

in their development, and are not regarded as invalidating the general conclusion.

Mating season.—Mature females to the number of twenty-eight were examined without finding sperm in the cloaca on the following dates: September 18, October 14, 17, 24, 31, November 5, 9, April 21, 23 and 26. Sperm have been found, on the other hand, in the cloacas of mature females on these dates in different years: October 31, November 5, 9, December 9, April 19, 21, 25 and 26.
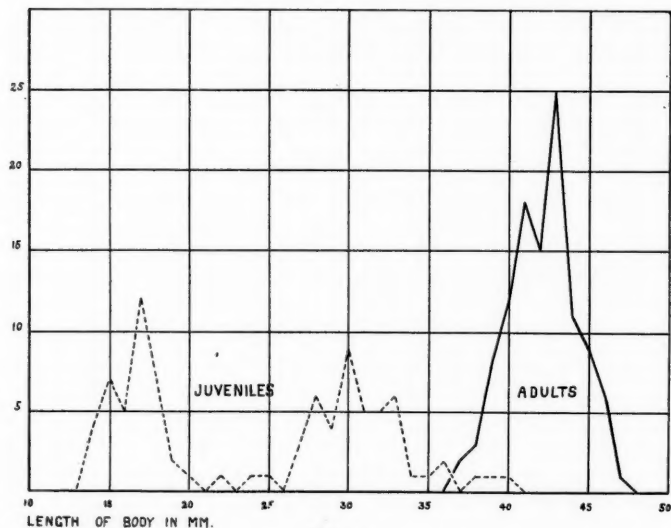

LENGTH OF BODY IN MM.

Fig. 4

This last set of dates is represented by thirty-four specimens, of which twenty-one were taken on October 31. On this date only six mature females were found without sperm. In other words, about two thirds of the sexually mature females collected on October 31 had already mated; but in thirteen specimens collected on earlier dates no sperm were found.

Males with the vasa deferentia crowded with sperm, which oozed freely into the cloaca in masses, were found on September 16, 18, October 24, December 9, April 17,

19, 21, 23, 26 and May 13. These dates are represented by twenty-seven individuals. Many other specimens were not dissected, but of those of adult size that were dissected within the limits of these dates only two had the vasa deferentia without sperm. These two represent the dates April 5 and May 25. The individual collected April 5 was one of only two of this species found on that date. Its stomach was empty and it had doubtless just emerged from hibernation. The testes of this specimen were full of sperm. The lack of sperm in the vasa deferentia of the specimen collected on May 25 may be accounted for by assuming that the mating season had passed.

These observations on both sexes indicate a prolonged mating season beginning near the end of October and extending through the first weeks of spring; but the autumn is indicated as the time when most of the mating takes place.

Spermatophores.—Spermatophores were first found December 9, 1923, in each of two females collected under leaves near together and in close proximity with four adult males. The spermatophores were pure white, subspherical bodies tucked closely into the posterior part of the cloaca. They resembled eggs of this species but were smaller. They seemed to be nothing more than sacs or bags filled with sperm. One of them measured one and a half millimeters in length and one and a quarter in width. One female with a spermatophore was found on November 5, 1924, and another four days later. Six more such females were obtained on October 31, 1926. Altogether ten females bearing spermatophores were taken on dates from October 31 to December 9. It is likely that the spermatophore is not retained longer in the cloaca than necessary for the spermathecae to be filled, for no shrunken spermatophoral sacs were found. Only in the first instance, described above, was any marked localization of adults of the two sexes noted. It seems probable that mating takes place wherever the males find unmated females and that there is no congregation of the sexes at

a special time or habitat, and furthermore that the spermatophores are directly transferred.

Dates of activity.—In common with other terrestrial salamanders of southern Michigan (species of Amby-stoma, Triturus and Hemidactylium) the red-backed sala-mander may be found early in the spring and late in the autumn. In the dried and hotter parts of the year it lives deeper in the ground and may not be easy to find. The writer's earliest spring date for finding this species is March 25 (1923) and his latest is December 9 (of the same year). Individuals had been feeding on both these dates and on the latter date females had fresh spermatophores. Breeding activity, however, as shown in the foregoing account is not an early spring function as it is with the Ambystomas of this region and many of the tailless amphibians, but is spread over several weeks of autumn and spring. Egg-laying probably occurs here in June, perhaps early to middle June, but the writer has no field data for this region. In the northern part of the lower peninsula of Michigan there is some evidence that the eggs are laid soon after the middle of June.

Food.—Examination of seventy stomachs from speci-mens taken at various dates in spring and autumn showed twenty-nine to contain beetles; nineteen, ants; seventeen, spiders; ten, small snails, mostly flat-coiled; six, millepeds; five, bugs; four, mites; three, springtails; three, pseudoscorpions; two, earthworms; and one each, weevil, sowbug, pupa, hymenopteron, maggots, roach, inchworm and amphipod. Various insect larvae were found in twenty-five stomachs, and miscellaneous uniden-tified small insects in thirty-four stomachs. This is prac-tically a verification of the findings of Surface (1913). It shows a preference for and ability to catch relatively large-sized and active forms, mostly insects, but also, prominently, spiders, snails and millepeds. Shed skins were found in twelve stomachs. How often the skin is shed and whether it is regularly eaten seem questions yet to be answered. In addition to the seventy individuals

whose stomachs contained food, eleven examined had empty stomachs. These represented various dates on which individuals with full stomachs were obtained. Most stomachs contained a variety of contents, but in a few cases a single form made up the bulk; the contents of one stomach, for example, was made up largely of a mass of black springtails, and another stomach was crowded with ants.

Summary

1. Mature males have a body length that is, on the average, a little shorter than that of mature females.

2. The red-backed and black-backed phases are approximately equally numerous in the vicinity of Ann Arbor, Michigan, and bear no relation to sex or age.

3. Total egg complements vary from five to thirteen, and the eggs are, on the whole, equally distributed between the two ovaries.

4. There appears to be no correspondence between the length of the female and the total number of eggs produced.

5. Breeding age is attained at the end of the third season, *i.e.,* at about two years of age, and the first eggs are laid in the spring of the fourth season.

6. The mating season is autumn and probably also early spring.

7. Spermatophores have been found in the cloacas of ten females, between the dates October 31 and December 9.

8. The species is active very early in the spring and very late in the autumn.

9. The food is largely spiders, beetles, ants, snails, insect larvae and various small insects.

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ULTRA–VIOLET AND EXTINCTION[1]

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EXTINCTIONS of the geologic past have attracted much study and have led to various speculations. The reader who turns from the baffling effort to appreciate the vast stretches of time between us and the countless exterminations recorded in the rocks must be surprised at the amount of fairly exact knowledge which has been assembled in explanation of those remote disasters. Indeed, it is not unusual in our own day to find that our efforts to understand the cause of death of an individual do not result in an explanation much more precise than that of the geologist who is making a post-mortem diagnosis on an entire ancient genus or family.

Osborn (ref. 1: ref. 2) divides the causes of extinction into the internal and hereditary factors and those which are external or environmental. Lowered racial vitality, deficient mentality, reduced rate of propagation, inadaptability and irreversible specialization of structure, are among the internal factors. Environmental causes of extinction range from stupendous movements producing mountains; include secular changes in climate and rainfall, and even such insignificant influences as the arrival of a new competitor for food; the disappearance of a food source, or invasion by a parasitic disease. There are, nevertheless, still many extinctions not clearly understood. For this discussion we may consider those occurring between Triassic and Jurassic; the mammalian extinctions of Pliocene time, and the devastation attend-

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ing the end of the Mesozoic and that of the great Glacial
Ages of Pleistocene.

It is the object of this paper to suggest that animal life
is distinctly influenced by either excesses or deficiencies
of ultra-violet rays from the sun; and, particularly, that
ultra-violet deficiency deserves consideration as a cause
of animal extinction at times when plants survived; to
suggest, further, that ultra-violet deficiency ought, in
general, to be more important as a cause of those extinc-
tions which were widely spread over the face of the earth,
which occurred at times of cool or cold climates, and
under conditions favoring selective reduction of the
shorter rays from the sun.

Section II
Geologic Summary

In Triassic America the continent was elevated and
inland seas were largely absent; the climate, though
cooler than Jurassic, was warm.   The red sedimentary
deposits indicate a shortage of rainfall.   While the rep-
tilian rulers apparently required much warmth and great
moisture, they nevertheless flourished through Triassic.
There were, however, local humid swampy regions.

During early Triassic there were active volcanoes from
Alaska southwards, which do not correlate with extinc-
tions.   During Upper Triassic, volcanoes around British
Columbia were very active, depositing as much as 10,000
feet of lava and ash (ref. 3, p. 467).

Triassic closed with the Palisade disturbance, a great
ridge being folded up along the Atlantic in eastern Amer-
ica.   With this there was a period of cooling, extending,
apparently, over a large part of the earth (ref. 3, p. 513).
In this interval, apparently, between Triassic and
Jurassic there was a sharp assault on animal life.   Sev-
eral orders of the earlier gigantic dinosaurs succumbed;
at least 2,600 species of ammonids disappeared from the
seas; there was a geographic restriction of reef corals,
and insects were dwarfed.   Plant life showed no signifi-
cant break, the ancient types—conifers, rush, cycad, fern

—continuing from Triassic into Jurassic (ref. 3: ref. 4: ref. 5: ref. 6: ref. 7, tables, pp. 53 and 79).

This, the earliest of the occurrences selected, is by no means a convincing case. The extinctions at this time offer only a suggestive example for our discussion, as it is possible that the interval between Triassic and Jurassic was not actually the period of massive extinction (ref. 4, p. 93; p. 549). The disappearance of the earlier great reptiles, instead of occurring at this interval, may have been a gradual process extending over late Triassic and a large part of Jurassic. However, after summarizing the data on this subject from the authorities quoted, the evidence seems suggestive enough to be included.

The cool temperature is apparently the chief reason assigned for all the faunal changes except those of the dinosaurs, and here, as additional causes, oversize, over-specialization and low mentality are brought in as factors.

It may be noted that no great temperature change, and no great extinction was associated with the two next great land risings during the age of Reptiles. Also volcanoes were active in this interval, so that it is obvious that vol-canoes can be active without producing extinction, and some further factor must be sought, possibly a difference in the intensity, or duration of volcanic output, or the nature of the smoke and dust. Nevertheless, it appears that between Triassic and Jurassic there was a correla-tion of extensive animal extinction, plant survival, cool temperature and volcanic activity. It is suggested that ultra-violet deficiency was a contributing factor in these extinctions.

In our next example, mountain-making on an enor-mous scale marked the "critical period" at the end of the Mesozoic Era. The Andes, Rockies and other mountains were formed; the Appalachians were re-elevated. The greatest known volcanic activity occurred in Africa and Arabia. The continents were high, inland seas receded, and there were extensive arid areas, or even

a general reduction in rainfall. The world climate became cooled, but no extensive glaciation seems to have occurred, though evidence has been found of local glaciers in Colorado. This cooling marked a sharp contrast to the prevailingly warm temperatures of Cretaceous and Eocene, which preceded and followed the "critical period." It seems possible to trace the geologic record in some places with no serious break from Cretaceous time, before the Critical Period, into the subsequent Eocene Epoch (ref. 8).

Extinction occurred at this time on a majestic scale, world-wide in extent, involving both land and marine creatures. Thirteen orders of reptiles were wiped out, both land dwellers and marine forms. Teethed birds disappeared. Entire races of molluscs were exterminated, including ammonids, belemnids and rudistids. There was a marked reduction of corals and ganoid fishes (ref. 3). A gradual decadence of the reptilian ruling line preceded the final crash (ref. 9, pp. 193, 194). Osborn speaks of this disaster as one of the most dramatic moments in the life history of the world, and says: "We have no conception as to what world-wide cause occurred" (for the dramatic exit of the reptilia). . . . It is natural to attribute this extinction to a general lowering of temperature but the flora shows no evidence of this either in Europe or America, nor is there evidence of any great geographic cataclysm on the surface of the earth, for the plant life transition from one age to the other in the Rocky Mountain region is altogether gradual and gentle (ref. 2, pp. 97–98).

There was no important extinction in the plant kingdom at this critical period. On the other hand, changes of great importance in plants preceded the vast destruction of animal life associated with the end of the Cretaceous. According to Berry (ref. 10, p. 398), the ancient plants which endured into the late Lower Cretaceous either disappeared or were very much reduced in importance in Upper Cretaceous, while the newer types, the

flowering plants, gained ascendancy.   The lines of descent
of most of our forest trees follow down from this period
of Upper Cretaceous (ref. 11, pp. 48, 51: ref. 5, p. 132).

It may be that dinosaur extinction depended more or
less on the gradual reduction of the older plants on which
they fed.   Possibly they could not adapt themselves to the
newer vegetation.   On the other hand, the warm-blooded
vertebrates which had remained for the centuries from
Upper Triassic an humble, undeveloped line, may have
found in this new vegetation that mysterious stimulus
which projected them, alone of animal types, upon a
career of evolution and made them masters of the world
(ref. 39: ref. 2, p. 82; p. 84).   Practically all the impor-
tant food plants are modern types.

Explanations of the tragedies marking the close of the
Mesozoic refer largely to the dinosaur extinctions.   Ap-
parently no suggestion has been made of any other uni-
versal or general cause than cooling as a common factor
in the destruction of both land and marine forms, large
and small.   May it not be that there was a common cause
contributing to the downfall of the extinct races, and, in-
directly, through influence on the plant world, preparing
foods needed for the active evolution of birds and mam-
mals?   It will be shown that ultra-violet deficiency may
be an important factor in the changes at this critical
period; that it offers a cause for world-wide extinctions,
and that the physical conditions indicate a reduction or
blocking out of the short rays of the sun at that time.

In our third example, the great glacial ages of the
Pleistocene epoch stopped the course of life for many
types of mammals.   The vast pendulum-swings between
the ages of ice and the centuries of nearly tropical cli-
mate caused waves of floral movements back and forth
from the warmer zones, and with the plant movements
there were animal migrations.   The combinations that
caused extinction must have been varied and numerous,
but exact conclusions are difficult.   Osborn decides that
proof is lacking of a *direct* connection between extinction

and the great geographic and climatic changes of the glacial times. Mentioning a number of *indirect* causes which contributed to the great losses of life, he concludes that the facts only justify a view that the Glacial Time in North America originated certain new conditions which directly or indirectly resulted in extinction (ref. 3, pp. 501–509). Active mountain-making and abundant volcanoes distinguish the Pleistocene. Fogs and clouds fringed the glaciers.

The glacial climates, as they advanced, killed back plant life; as the glaciers melted plants and trees grew up in the old glacier paths. Post-glacial conditions activated plant evolution especially among herbaceous forms (ref. 10, pp. 406–407).

That ultra-violet deficiency may have contributed to the Pleistocene devastation is suggested by the cooling of the atmosphere to the point of glaciation; by the evidence of intense volcanic activity, and by the extent of the losses sustained. The plant losses are explainable as directly due to cold and glacial conditions. In the neighborhood of glaciers fogs and clouds would still further screen the earth from the sun's short rays.

One period of expansive evolution suggests an ultra-violet influence, contrasting with the three great extinctions described above. The evolution of mammals was at its height during the Miocene Epoch and into the Pliocene and early Pleistocene. A distinct reduction occurred in the ancient types of plant (fern, cycads, etc.), with a further spread of the flowering plants, and the first general use of grasses as food; a food rich in vitamines (ref. 2, p. 94). While migrations, new enemies, climate and other causes were important in the many extinctions during this interval, the abundance of volcanic ash, and the evidences of cooling point toward selective alterations in ultra-violet radiation as having also had a part in the biologic changes. It was during the epochs of this era that the greatest evolution occurred in teeth and in certain bones, such as those

of the foot.   There are close relationships between the growth of bones or teeth, and the ability of the body to use lime or calcium and phosphorus; close relationships also between the "calcium balance" and "phosphorus balance" of the body and a sufficiency of ultra-violet radiation; close relationships also between animal growth, calcium balance and the plant vitamines dependent on ultra-violet rays in the sunlight.   It may be surmised, in view of what is known of the processes of evolution, that the tooth and bone developments in this era were reciprocal to changes in environment which in some way stimulated the calcium activity of the genera concerned.   It may be surmised that ups and downs in ultra-violet dosage depending on the amount of volcanic dust, with a corresponding influence on plant vitamines was one of the stimuli to evolution, the inadaptable forms perishing (ref. 2, pp. 369–370).

To sum up: Three points are selected out of all geologic time: (1) That extending from the latter part of Triassic to the **early** Jurassic; (2) the "critical period" at the end of the Mesozoic, covering the period of change from Cretaceous to Eocene; (3) the Pleistocene critical period.   In each instance there is evidence of extensive animal extinction, wide-spread, involving many species; absence of plant losses of any significance; evidence of abundant volcanic action; the record of a cooled climate —a combination indicating ultra-violet reduction.   For contrast, the period of mammalian expansion is selected as suggesting that fluctuation in sunlight, with volcanic dust interruptions, influenced mammalian evolution by favoring virile species with the more plastic calcium mechanisms, and thus was moulded the new foot-and-tooth structures of that period.

SECTION III

ULTRA-VIOLET

Sun's rays of wave-length shorter than approximately 400 millimicrons are invisible, the "ultra-violet rays."

As the wave-length approaches 300 millimicrons the spectrum rapidly decreases, then ends abruptly (ref. 12, pp. 3, 19, 24). Little is known of variations in the output of solar energy, aside from sun-spot changes, and other slight variations with rather irregular cyclic recurrences (ref. 13, pp. 560–561). Part of the solar energy is absorbed in transit from sun to earth. The sudden ending of the solar spectrum is apparently due to absorption of the shorter rays in the atmosphere. The atmosphere is progressively less transparent toward the shorter wave-lengths, with increasing molecular scattering of light. The evidence indicates that in the dry upper atmosphere, above 40 kilometers, wave-lengths of less than 200 millimicrons convert oxygen into ozone which absorbs the shorter rays, and that longer rays and moisture dissociate ozone. In addition oxygen nearer the earth absorbs rays shorter than 366 millimicrons. It seems to be established that ozone in the upper atmosphere and oxygen are responsible for the abrupt ending of the solar spectrum (ref 12, Chapter II).

This may be a fact of some theoretic importance. The shorter ultra-violet rays, produced experimentally, are more destructive to life than the longer ones. Bayne-Jones finds the bactericidal action of light begins at 350 millimicrons and becomes progressively more intense to 185.6—the limit of his measurements (ref. 14, p. 11). If there was ever a time when the earth's atmosphere was low in oxygen, it would have been a time both unfavorable for respiration and with unfavorable radiations (ref. 15: ref. 12: ref. 16, pp. 239–66).

Water vapor intercepts ultra-violet rays to only a slight extent. Water vapor has an absorption band maximum near 175 millimicrons, but this field is not satisfactorily explored (ref. 12, p. 39). Carbon dioxide has absorption bands in the extreme ultra-violet, but it and nitrogen are practically transparent to the middle ultra-violet (ref. 12, pp. 31, 39). The condensed moisture in clouds and fog is a great barrier to ultra-violet as to other rays.

According to Humphreys, ultra-violet output from the sun is influenced by sun-spots; during sun-spot maxima the spectrum is poorer in ultra-violet, and richer during sun-spot minima, but the total effect is not very great (ref. 13). Brooks (ref. 17) quotes a suggestion from Humphreys that cosmic dust may intercept sun's rays to some extent.

While ultra-violet rays may be reduced by oxygen, ozone and sun-spots, it is probable that the most important factor in this connection is volcanic dust. This subject is fully developed by Humphreys (ref. 13), who presents convincing evidence that, under certain conditions, volcanic dust intercepts part of the sun's rays. He advances the theory that volcanic dust has been an important factor in the production of some of the glaciers. He gives detailed discussion of the sizes of volcanic dust particles; the distance they are projected into the upper atmosphere; their period of suspension, and the reduction in radiation due to the refraction, reflection and diffraction of sun's rays, and establishes the proposition that volcanic dust intercepts the solar rays to a notable extent. At the same time the dust particles interfere much less effectively with the loss of heat energy from the surface of the earth, the wave-lengths from the earth's surface being longer.

On these two conceptions, namely, the interference by dust clouds with the arrival of solar energy, and the almost unaltered dissipation of heat from the earth, Humphreys bases his theory of the cause of glaciation.

The theories to account for glaciers are discussed in text-books of geology and in recent works by Brooks (ref. 17) and W. B. Scott (ref. 18). There are, possibly, several factors involved in glacier formation. It is not the object of this paper to discuss glaciation nor to defend any theory of glacier formation. If the arguments presented here are sound, they may well bring support of a biologic nature to Humphreys' theory. If that part of Humphreys' work is not correct in which he traces out

the intercepting action upon the sun's rays of volcanic
dust and of sun-spots, the argument of this paper is
greatly weakened.   On the other hand, the validity of
this thesis does not depend upon the truth of the theory
that dust actually causes glaciers.   The shorter wave-
lengths from the sun may be lost to the earth with a rela-
tively slight reduction of the sun's heat.

Humphreys notes, as incidental to his main theme, that
suspended volcanic dust particles do not intercept all
rays from the sun with equal completeness.   The shorter
wave-lengths are more completely blocked than the
longer ones.   In fact, the interference by a particular
size of dust particle will vary inversely as the fourth
power of the wave-length, so that light in the extreme
violet end of the spectrum will suffer about sixteen
times as much interference as the rays at the red end.
In other words, the extremely small particles of volcanic
dust have a partially selective action on the sun's rays,
interfering particularly with the shorter wave-lengths.
Humphreys estimates that the volcanic dust particles
may require as much as one to three years to settle to
the earth from the upper atmosphere after the cessation
of volcanic action.   Prolonged volcanic action or erup-
tions repeated at intervals of a year or two would nat-
urally prolong the dust effect over periods corresponding
to the duration of volcanic activity.   The dust from a
large eruption quickly envelops the whole earth, spread-
ing out in the higher levels of the atmosphere.

If Humphreys correctly presents the relations between
volcanic dust and ultra-violet radiation, we may say that
ultra-violet radiation to the earth from the sun decreases
during periods of atmospheric dustiness and increases
when the upper atmosphere is dustless.   Moreover, the
reduction in ultra-violet is disproportionately great as
compared with the cooling of the earth's surface from the
same cause.   A fall of even a few degrees in the earth's
temperature would correspond to an almost complete
removal of ultra-violet radiation to the earth from the

sun. Moreover, if Humphreys' estimate is correct of the
time which it takes for dust particles to settle from the
upper atmosphere, the very small particles which will
interfere with ultra-violet rays will settle more slowly
than the larger ones and, therefore, the effect of volcanic
dust in reducing the ultra-violet radiation will not only
be more pronounced than its cooling effect but will last
longer.

Section IV

Ultra-violet and Plant Life

Palladin (ref. 19) states that sunlight plays a part in
the formation of chlorophyl; in its specific functioning,
and in plant growth. A few very ancient plants can form
chlorophyl even in the absence of light (conifer seedlings,
fern fronds, some one-celled algae). Even these do bet-
ter in the presence of light, and other plants require light
for the formation of chlorophyl. Very weak light is suf-
ficient; light of medium intensity is best. In light of
weak intensity, chlorophyl is formed faster under the
red-green rays. In the presence of filtered ultra-violet
radiations greening is very slight. In other words, re-
duction of the ultra-violet radiation will have no appre-
ciable effect on the formation of chlorophyl.

Chlorophyl absorbs most completely the wave-lengths
between B and C of the spectrum; that is, those at the
yellow end (wave-lengths of 650–666 millimicrons) and
these are of most importance in photosynthesis. How-
ever, the amount of carbon assimilation by chlorophyl is
independent of wave-length and, within limits, is directly
proportional to the amount of energy absorbed by the
leaf. The greater the amount of chlorophyl the less is
the amount of light required for photosynthesis.

Spoehr (ref. 20) in general agrees with Palladin. The
intensity of light is more important than the wave-length
(p. 110). Ultra-violet will not induce starch formation in
leaves, a process more complex than simple CO_2 absorp-
tion (p. 111). Photosynthesis becomes less with shorter

wave-lengths (p. 112). Whether ultra-violet rays alone induce photosynthesis is uncertain and is complicated by the injurious effect of those rays on the enzymes and other constituents of the plant (p. 118).

The growth of plants is retarded by ultra-violet radiation, but not by yellow rays. The greater the growth-retarding effect of light, the stronger is its phototropic influence. Plants require the blue-violet rays in order to grow in their usual forms (ref. 19, pp. 275–283). Plants require light for the formation of lipochrome and vitamine A, but ultra-violet is not necessary. The vitamine may be formed even in the absence of chlorophyl (ref. 21, p. 134).

However, Luckiesch advises that it is unsafe to be too positive in statements as to the effect of ultra-violet radiation on plants (ref. 12, p. 219). More systematic and accurate work will be required before specified wave-lengths can be correlated with definite plant reactions. It will probably be found that some species will be more sensitive than others; and that differences in dosage of rays will yield diversities in result. Schanz states that plant albumen is sensitive to radiations of short wave-length, while chlorophyl is brought about chiefly by longer wave-lengths: also that plants protected by plain glass or by Euphos glass grew larger than those exposed directly to the sun. The plants completely protected from the ultra-violet and blue rays were much larger, and, though green, reminded him of etiolated plants (ref. 22). Palladin and others also connect etiolation in plants with ultra-violet deficiency, and, furthermore, there is other evidence that ultra-violet may inhibit plant growth, and has a tropic influence.

This review shows that information on the subject is not complete nor consistent, but makes it seem highly probable that reduction at the ultra-violet end of the sun's rays, from dust in the upper atmosphere or other cause, may affect the forms of plants but will apparently have no serious effect on the formation or activity of

chlorophyl in those plants surviving to modern times. Plants growing in sunlight poor in ultra-violet might be larger than the average for their species; and under certain obscure conditions may be etiolated.

Section V

Ultra-violet and Animal Life

When it reaches the earth, sunlight does not contain the very short wave-lengths which emanate from the mercury arc. Pure water is quite transparent to the near and middle regions of ultra-violet radiation and is opaque to infrared. Only about one per cent. of solar energy penetrates as deep as four meters into water, the rest being absorbed (ref. 12, p. 31). Dilute solutions of various salts reduce the transparency of water for ultra-violet (ref. 12, p. 48). Rays of a wave-length of 487 millimicrons may go to a great depth (ref. 12, p. 50). Ultra-violet radiation has been detected at 1,000 meters depth of ocean, but not at 1,700 meters (ref. 12, p. 217). Radiation needed for vegetation does not extend below 400 to 600 meters.

There is a most abundant life floating in the upper levels of the ocean which is an important source of food for marine creatures. It is not known, however, whether the sea forms are dependent on ultra-violet, or, on the other hand, are injured by those rays. Small crustacea flee from ultra-violet radiation. The rays may be toxic or lethal for bacteria and protozoa. The shorter the wave-length the more lethal is its action on minute creatures. The lethal dosage is largely of wave-length shorter than that received from the sun. The marine plankton is more abundant in winter than summer waters; and in foggy regions more than in clear (ref. 12, p. 218). Marine life with fixed habitat is generally established in the levels reached by the sun's rays. In the absence of knowledge on this subject, it is here suggested that marine animals resemble land animals in requiring ultra-violet either as sunlight received directly upon the body or in the form of irradiated food.

Ultra-violet irradiation may exert in land animals either a stimulating or irritating effect, depending on the dosage and wave-length. Pathologic effects from the action of ultra-violet are known in human beings, such as sunburn and certain eye diseases.

Of greater importance are the recently discovered relations between ultra-violet and general nutrition. Nutrition can be improved in some undernourished human beings by exposure to sunlight or ultra-violet rays in controlled amounts. The metabolic rate is increased, lymphocytosis is produced and old resisting infections are in some cases more successfully opposed by the patients (ref. 23: ref. 15). It is not possible to attribute all of the good effects of sun-baths to ultra-violet rays. Sun baths are more beneficial than ultra-violet baths alone in many cases. The rays with less refrangibility penetrate deeper (ref. 30: ref. 40).

It has also been found that hens exposed to ultra-violet lay regularly all winter; the eggs have a higher percentage of hatchability and the hatched chicks possess greater vitality than the offspring of other fowls not exposed to the ultra-violet rays (ref. 24: ref. 25).[2]

Other creatures, also, including some of the present-day reptilia, require ultra-violet rays. It is stated that "ultra-violet rays have a markedly beneficial influence on the vitality and spirits of reptiles, monkeys and the large carnivores." In connection with the regular use of powerful ultra-violet lamps at the London Zoological Gardens, "delicate monkeys that formerly moped all winter were seen basking in the light rays and dashing in and out of the cages," . . . "delicate reptiles that formerly never ate and seldom survived the winter were seen eating vegetable food with relish" (ref. 28).

Ultra-violet deficiency has been shown to have a close connection with serious disorders of man and experi-

[2] Investigations are reported in recent journals on special vitamines controlling sterility and fertility, and on whether poultry are like other animals in this regard (ref. 26: ref. 27).

mental animal. Rickets (rachitis) and tetany are both intimately connected with this cause. The calcium or the phosphorus in the blood serum is reduced, or calcium-phosphorus ratio is disturbed (ref. 29: ref. 26). Tetany is seen in very young infants and is apt to end fatally with convulsions. Rickets, a disease of growing children or young animals, is characterized by nutritional and growth defects, by anemia, digestive disorders and lowered resistance to infectious diseases. In severe cases it resembles scurvy. It is often the predisposing factor in a fatal illness. Experimental rickets in adult animals is characterized in its milder forms by hardly any other change except sterility or loss of fecundity, or reduced vitality of offspring. If the diet is properly selected, ultra-violet irradiation of rachitic children or animals stops the progress of the disease and effects a cure by restoring the defective calcium or phosphorus balance in the blood and permitting of a more economical use of these constituents of the body. In the cure or prevention of rickets the patient may be treated either directly with sunlight or the ultra-violet lamp, or he may be given the vitamine in his diet, or dosed with cod-liver oil. Apparently he receives the ultra-violet energy directly from the sun, or its equivalent indirectly with his food, while cod liver oil seems to represent at least two removes between the sun's rays and the patient; first, the irradiated food eaten by the cod fish; second, the cod's liver, from which the oil is expressed for medicinal purposes.

It has been recently stated that experimental rickets can be prevented and cured by feeding grasses and fodders which have grown or been cured in sunlight and have stored the antirachitic property or substance. It has further been noted that winter-grown grasses and spinach are lacking in power to prevent rickets—a fact apparently connected with the ultra-violet shortage of winter sunlight (ref. 32: ref. 33: ref. 34). It has, moreover, been possible to confer upon certain plants the power to protect against rickets by treating them with

the ultra-violet lamp (ref. 35).   Also Hess and others
have shown that certain oils, after irradiation with the
ultra-violet lamp, carry the power to cure or prevent rick-
ets (ref. 36: ref. 37).   It appears from reports by Hess
and Shipley (ref. 26) that wave-lengths between 210 and
334 millimicrons are the ones chiefly active in curing and
preventing rickets.   As the solar spectrum ends abruptly
at wave-lengths between 290 and 300 millimicrons, the
antirachitic solar rays would seem to occur only in the
narrow range between 290 and 334 millimicrons.   These
observations have a possible bearing on some of the ex-
tinctions, for modern grasses and cereals could hardly
have been abundant before the Miocene or Pliocene
epochs, though they took their start before the fall of the
reptilian dynasty.   If the older types of fodder; fern,
cycad, etc., could not store the antirachitic property, the
animals of that day would have been more dependent
upon direct exposure to the sun for this property than is
true of modern animals.

Bayliss (ref. 15) and Spoehr (ref. 20) discuss the
mechanism of radiant action, but ultra-violet mechanism
is obscure.   The rays are absorbed at the surface of the
body, and, experimentally, by blood serum, spinal fluid
and various tissues.   Irradiated serum shows an increase
of dialyzable calcium (ref. 31); irradiated plasma takes
up oxygen: irradiated blood takes up oxygen at low oxy-
gen tension, and loses it at high, with equilibrium at 15
mm. of oxygen.   A large number of proteins and trypto-
phane and tyrosine take up oxygen under the ultra-violet
lamp (ref. 41).   Ultra-violet rays alter the globulin con-
tent of egg albumen (ref. 42), and ''sensitize'' egg albu-
men and the tissues of the protozoa, ''Paramecia,'' so
that coagulation occurs at lower temperatures than
before irradiation (ref. 43: ref. 44).   As mentioned above
ultra-violet rays injure plant proteins and destroy bac-
teria and protozoa.

It is, however, questionable whether the results of
recent experiments can be applied to the geologic prob-

lem. Most of the experiments have been conducted with carbon, fluorite or mercury lamps giving abundance of short ultra-violet waves, which are lacking in the sunlight reaching the earth. There is also confusing suggestion that long and short waves may either supplement or block each other. The geologic questions are, then, speculative and suggestive, but, nevertheless, the established facts associating ultra-violet rays with health, and their shortage with tetany and rickets are beyond doubt.

Section VI

Discussion and Summary

According to the facts assembled in the preceding sections ultra-violet rays from the sun are necessary to the well-being of man and of the higher animal types thus far observed. Exposure to ultra-violet rays of the sun or of a special lamp has a decidedly beneficial effect in a number of diseased conditions. Deficiency of ultra-violet contributes largely to the causation of tetany and rickets, diseases which may have a distinct importance in preventing racial survival. Fertility of at least some animals diminishes or disappears after prolonged absence of ultra-violet rays. Many of the beneficial effects resulting from the ultra-violet rays of the sun playing directly upon the body can be obtained indirectly, by utilizing for food either animals properly irradiated or certain grasses or oils previously irradiated either by sunlight or an ultra-violet lamp. It is not yet possible to say how general is the dependence of animal types upon ultra-violet rays, nor to say what plants are capable of converting the ultra-violet rays into stored antirachitic property. Winter sun does not give good ultra-violet irradiation to plants. Summer sun does.

It has been shown that plants are not apt to suffer serious damage in the absence of the short rays of the sun. Though their value as food is lessened, survival would not be endangered.

Several causes have been mentioned which alter the amount of the sun's ultra-violet reaching the earth and,

in particular, Humphreys' analysis of the effect of volcanic dust upon the sunlight has been reviewed. If Humphreys' determinations are correct, it follows that volcanic dust sufficient to cause slight cooling would cut off practically all ultra-violet rays from the earth. Under conditions of prolonged, intense volcanic action, no ultra-violet rays would reach the earth for, possibly, years at a time.

The geologic record indicates that there have been periods of prolonged intense volcanic action. On some of these occasions there was no recorded cooling of the earth, and no recorded animal debacle. This can not be explained. On other occasions, it appears that there was a concurrence of volcanic action, a cool or cold period, tremendous animal mortality and no extinction of plants. This is exactly the combination to be expected if Humphreys' views are correct and if prehistoric animals were as dependent as modern animals upon ultra-violet rays.

If the ancient types of animal were dependent upon the sun's short rays, ultra-violet deficiency should have been followed by rather rapid extinction. When the cold-blooded reptilia and other animals which flourished through the many warm centuries of the Cretaceous were deprived of their accustomed warmth, as the world's climate gradually cooled, either they migrated or they must have become more and more torpid and helpless. Their metabolic activities and internal vital processes were at a lowered level. These conditions restricted the abundant cross-fertilizations which are favorable to the appearance of variant offspring. The sluggish creatures could not protect themselves or their eggs or offspring from enemies more active. The shorter warm seasons and colder climate caused the loss of many eggs and many of the young. The adults and young were probably forced by hunger to eat many strange plants, in the absence of accustomed fodder. Malnutrition and digestive disturbances followed the unfamiliar diets, conditions suitable for the production of rickets.

Favored by centuries of warmth and sunshine, the reptilia and other genera and species were faced with a cool temperature, the results of which have just been outlined, and, at the same time, with ultra-violet deficiency. Little or no ultra-violet radiation would reach the bodies of the creatures directly; the ferns and other fodders, also lacking ultra-violet energy, would be comparable to the winter grasses and winter-grown spinach which recent investigators have reported to be lacking in anti-rachitic property.

With their accustomed diet reduced by the effects of cold on plant and animal life; undernourished, torpid, and lacking the ultra-violet energy, the creatures would become less fertile, at the time that they could less successfully protect eggs and offspring, and at the time also that ultra-violet deficiency reduced the hatchability of eggs and lowered the vitality of the hatched young. The young, in their turn, with inadequate food during the growing periods, and with ultra-violet deficiency, would have the conditions causative of tetany or rickets. The unsuitability of the diet in conjunction with the digestive disturbances of rickets would be disastrous to the affected young. Their resistance to infection, lowered during rickets, would synchronize with a period in which the bacteria and protozoa would be released from the inhibiting influences imposed on them in days when ultra-violet rays prevailed.

The continuance for a few generations of the combination of factors reviewed above, in which ultra-violet deficiency is of importance, would be sufficient to cause the death and disappearance of the animals dependent on a warm climate and abundant ultra-violet radiation. Moreover, such a disappearance ought to occur rapidly— within a few generations—if the species is vitally dependent upon ultra-violet radiation, and if the reduction of that spectral region is pronounced. Extinction produced in this manner should leave only slight traces in the fossil bed. At the most, there would be the chance finding

of an enlarged rachitic bone, or of defective teeth, or of one of the rachitic deformities' of skull, pelvis or ribs (ref. 38: ref. 26).

Under the conditions reviewed, ultra-violet deficiency would usually be a cause of world-wide extinction.

Summary

In summary, it is here suggested that if dust between earth and sun and abundant sun-spots, separately or together, produce a cooling of the earth's surface, they will, to a greater degree, produce a selective reduction in the ultra-violet radiation from the sun to the earth. It is suggested that ultra-violet deficiency, which is associated with disease among some forms living in the present time, was an equally important causative factor in disease in geologic time. It is suggested that the disease features of modern tetany, and especially of rickets— sterility in the experimental adult animal, digestive, nervous and metabolic disturbance, anemia and lowered resistance to infection—were probably as severe, or even more severe in the earlier periods before species adaptation occurred. It is suggested that cold-blooded animals, chilled by a drop in temperature, were too sluggish and helpless to search for vitamine-containing foods, and their vital processes were so slowed down as to reduce the rate of evolution of new adaptations. It is pointed out that ultra-violet deficiency has relatively slight influence on plants, though possibly influencing their size and form, and is more destructive to animals, which fits in with the evidence that the wide-spread extinctions at the end of the Cretaceous were confined to animals, while plants continued to evolve at a steady rate. The same combination between Triassic and Jurassic—cooling, volcanic ash, wide-spread animal extermination, but no plant extinctions—indicates that ultra-violet deficiency was the common factor in this destruction, though in this case the evidence is weak.

It is suggested that intermittent volcanic action during the Miocene and Pliocene epochs, leading to periods of

ultra-violet deficiency, was one of the causes of the extinctions recorded in the fossil beds of Western America, and stimulated a reciprocal protective evolution on the part of surviving forms.   There may have been a connection between this evolutionary process and the spread of the flowering plants.   It is at least not improbable that surviving species have developed a wider range of adaptability to variations in the ultra-violet radiations than was possessed by earlier progenitors.   Survival of ultra-violet deficiency in this generation with the pathologic reaction "rachitis" may represent a partly developed adaptation evolved by ancestors in a distant geologic period under the pressure of ultra-violet deficiency.   Von Hansemann's estimate (ref. 29), that rickets is exclusively a disease of human beings and their subjugated animals, may be true to-day, but may need revision if a longer view is taken into the past.

Since much the same combination of cooling, volcanic action, animal destruction and plant survival occurred at the Pleistocene, it is probable that ultra-violet deficiency contributed to the destruction at that time.

It is suggested, finally, that selective variation in the intensity of rays from the several parts of the solar spectrum may be a factor in extinction; and that ultra-violet deficiency may be an example of such a phenomenon. With protracted, severe ultra-violet deficiency there should be little or no extinction of plants, but animal extinction should be wide-spread over the face of the earth: should involve many animal types, and the extinctions should be completed within a geologically brief period.

It may be added, as a further conclusion from the data assembled in this study, that the relative intensity or quantity of ultra-violet radiation in different latitudes from the equator to the polar regions deserves to be investigated as one of the possible factors influencing the distribution of fauna.

It may be possible to establish or disprove the validity of this thesis by examination of fossil bones and teeth, if

methods can be devised for the recognition of fossilized rachitic bones (ref. 26: ref. 38). From another point of view, a study of the distribution of rickets and its correlation with atmospheric haziness, or of volcanic dust, might furnish information bearing on this subject.

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SHORTER ARTICLES AND DISCUSSION

ORIGIN OF A SEGREGATE RESISTANT TO BLACK-STEM RUST IN A CROSS BETWEEN TWO SUSCEPTIBLE PARENTS[1]

HAYES, Aamodt, and Stevenson (1) have recently reported that from a cross of Minard x Minhardi they obtained some segregates that were semi-resistant to black-stem rust when both parents were susceptible. Their conclusion is: "Recombination of genetic factors evidently occurs in these winter wheat crosses whereby a cross of two susceptible varieties leads to the production of semi-resistant type." They obtained several such segregates.

Segregates showing the full resistance of the "R" type had been obtained by the writer during 1924 and 1925 in a cross between Dicklow and Sevier. Dicklow is completely susceptible; some pure lines of Sevier are completely susceptible, and some show varying amounts of semi-resistance. The writer was of the opinion that these resistant segregates had originated by transgressive segregation rather than having come directly from a fully resistant parent. Since, however, the pure line of Sevier from which these were obtained was not known, the cases were not as definite as was desired.

A result of considerable importance has been obtained recently in the plant-breeding work at the Utah Experiment Station in various crosses between pure lines of Sevier and pure lines of Federation (a completely susceptible variety). Homozygous segregates have been obtained, some of which show a semi-resistant (SR) reaction to black-stem rust and others of which give a reaction of the resistant (R) type. The segregates were produced at Logan, Utah, but the tests for rust were made in the artificial rust inoculation nursery at University Farm, St. Paul, Minnesota.[2] These tests were conducted during the two growing seasons 1926 and 1927.

[1] Contribution from the Department of Agronomy, Utah Agricultural Experiment Station.

[2] The rust breeding nurseries at University Farm, St. Paul, Minnesota, are under the joint direction of H. K. Hayes, head of the Section of Plant Genetics; E. C. Stakman, head of the Section of Plant Pathology, Minnesota Agricultural Experiment Station, and O. S. Aamodt, Associate Pathologist, Office of Cereal Crops and Diseases, U. S. Department of Agriculture. The writer gratefully acknowledges the courtesy extended of making the rust tests and supplying the readings of the same.

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The 1926 test showed one segregate to have the R type of resistance, several of the others being of the SR type. Table I presents data which prove that out of 81 segregates tested in 1926, ten were semi-resistant (SR) and one was resistant (R). One of the semi-resistant segregates and the only resistant one obtained that year came from a cross where the Sevier pure line was not known. The data on these two segregates are given

TABLE I

Percentage infection of black-stem rust on Federation, on pure lines of Sevier, and on homozygous segregates from crosses of Federation x Sevier. (Grown at St. Paul, Minnesota, 1926.)

Strain	Fully Susceptible (S)	Semi-Resistant (SR)	Fully Resistant (R)
Federation	90		
Sevier No. 60	80		
Sevier No. 101	90		
Dicklow	90		
Hard Federation	75		
Federation x Sevier No. 60			
No. of Segregates	5	2	
Percentage Range	75–90	60–75	
Federation x Sevier No. 101			
No. of Segregates	5		
Percentage Range	75–90		
Federation x Sevier No. 59			
No. of Segregates	50	7	
Percentage Range	50–95	40–65	
Federation x Sevier No. ?			
No. of Segregates	7	1	1
Percentage Range	60–90	45	40
Dicklow x Hard Federation			
No. of Segregates	8		
Percentage Range	70–90		

in the next to the last line in Table I. Unfortunately, the parent plant from which this segregate with R type of resistance originated was not known, and on that account it could not be certain that the parent plant itself did not possess the R type of resistance, though such plants have not as yet been observed in Sevier. Therefore, in 1927, a rather large number of segregates of definitely known descent, together with their parental strains were exposed to artificial infection in the rust nursery at Minne-

sota. This time a segregate possessing the R type of resistance was obtained, and it was demonstrated that one parent was fully susceptible and the other possessed only a slight amount of resistance of the SR type. The data for 1927 are given in Table II.

TABLE II

Percentage infection of black-stem rust on parental strains of wheat and on segregates which give a reaction of no resistance (S) or which give the semi-resistant (SR) of the resistant (R) type of reaction. The percentage infection with leaf rust is also given for the segregate classified into the S, SR, and R classes, according to their reaction to black-stem rust. (Grown at St. Paul, Minnesota, 1927.)

Variety of Strain	S		SR		R	
	Stem Rust	Leaf Rust	Stem Rust	Leaf Rust	Stem Rust	Leaf Rust
Federation	100	85				
Sevier	75	70				
Marquis	85	65				
Sevier No. 59			75	75		
Sevier No. 60			65	65		
Sevier No. 101	70	70				
Federation x Sevier No. 60						
No. of Segregates	34	34	12	12		
Range	50–100	25–85	25–90	35–70		
Mean	83.5	60.4	46.7	52.5		
Federation x Sevier No. 101						
No. of Segregates	19	19	1	1		
Range	50–100	25–90	60	90		
Mean	85.5	60.5	60	90		
Federation x Sevier No. 59						
No. of Segregates	13	13	4	4	1	1
Range	70–90	50–95	50–65	50–75	25	40
Mean	82.3	74.6	57.5	65.0	25	40

In 1927, 84 segregates were tested. Of these, 17 were semi-resistant (SR) and one was highly resistant (25R). The reaction of both parents of the resistant segregate was also obtained at the same time. One of the parental strains gave somewhat different reactions to black-stem rust in 1926 and in 1927. Federation was completely susceptible (90 S and 100 S) both years. Sevier No. 101 was also completely susceptible both years (90 S and 70 S), but Sevier No. 60 which gave a reaction

of complete susceptibility (80 S) in 1926 was semi-resistant (65 SR) in 1927. Sevier No. 59 was not included in 1926, but in 1927 it gave a reaction for semi-resistance (75SR). A certain amount of such variation must be expected of strains which are faintly resistant or which are resistant to only one or two rust forms. Many of the 188 pure lines of Sevier studied at the Utah Station between 1919 and 1927 show partial but never high resistance to the physiologic forms of black-stem rust that occasionally attack wheat in that region.

The data for both 1926 and 1927 give with considerable frequency recombination of genetic factors as a result of which segregates are produced which are more resistant to black-stem rust than are either of the parental lines. This tends to substantiate the opinion already mentioned that resistant lines previously obtained (2) arose by transgressive segregation. It also corresponds to data reported in the Minard x Minhardi cross (1). Here, however, is a definite and important case in which a segregate having a really resistant reaction (25 R) was obtained from a cross in which one parent was fully susceptible (100 S) and the other only slightly semi-resistant (75 SR). This is clear-cut evidence that highly important recombinations of considerable magnitude do occur in the genetic factors having to do with the inheritance of reaction to black-stem rust, whereby resistant lines are obtained from crosses of susceptible lines. Incidentally, also, these cases furnish additional evidence of the complexity of plant genetics.

The percentage of infection of leaf rust is not very different in the three groups of segregates classified according to their reaction to black-stem rust. Table II shows the percentage of leaf rust to be fully as high as or higher in the susceptible (S) group than in the semi-resistant (SR) group. The only obtained segregate of the resistant type (25 R) also showed a relatively low percentage of leaf rust (40) as compared with the segregates in the other groups which were infected to the extent of from 25 to 95 per cent., with mean infections around 50 to 75 per cent.

The evidences of correlation between resistance to black-stem rust and susceptibility to leaf rust found in previous studies (1) are lacking here.

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METCALF ON THE BELL–TOADS AND THEIR OPALINID PARASITES: A CORRECTION

In my paper, ''The Bell-Toads and their Opalinid Parasites,'' published in the January-February number of the AMERICAN NATURALIST, this year, I omitted mention of the Philippine Bell-Toad, *Barbourula busangensis* Taylor & Noble.[1]   I have not seen this species or its Opalinid parasites, and having no notes of my own upon it and having misfiled Taylor and Noble's paper, I failed to include this species in the discussion. *Barbourula* apparently reached the Philippines *via* the East Indies, being an early northward wanderer from the southeastward migration which carried the ancestors of the Liopelmas *via* Australia to New Zealand.   Study of its Opalinids would be of interest.

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[1] Taylor and Noble, A New Genus of Discoglossidae from the Philippine Islands,—American Museum Novitates, No. 121, June 23, 1924.